PROCEEDINGS
OF THE
NATIONAL ACADEMY OF SCIENCES
Volume 31 January 15, 1945 Number 1
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THE AMOUNTS OF ACETYLCHOLINE IN THE DARK SKIN AND IN THE PALE SKIN OF THE CATFISH*

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Communicated November 24, 1944

The dark and the pale phases of the common catfish, *Ameiurus nebulosus*, are brought about by the activity of a single kind of integumentary color-cell, the melanophore, which is innervated by two sets of autonomic nerve-fibres (Parker, 1934). Preliminary evidence has been advanced to show that these two sets of fibres, like those associated with the vertebrate heart, are in one case adrenergic and in the other cholinergic. By the use of organic indicators the pale skin of the catfish has been shown to contain adrenaline and the dark skin acetylcholine (Ach), and by a determination made in the Harvard Laboratories the dark skin of the catfish was found to contain 0.078 gamma of Ach per gram of moist skin (Parker, 1940). At the same time that this determination was being worked out at Harvard three Chinese physiologists, Chang, Hsieh and Lu (1939), published a similar determination made on the skin of the Chinese snake-fish by the same method as was being used on the catfish, a determination which yielded 0.077 gamma of Ach per gram of skin. The very close agreement between the figures for these two determinations must be in a measure accidental. Nevertheless their agreement indicates beyond a doubt the approximate order of magnitude of the amount of Ach involved and shows that this amount is very small.

Ach is now well known to darken many fishes by inducing in their melanophores a dispersion of pigment. Presumably extremely small amounts of this agent are normally released from cholinergic nerves in effecting this dispersion. This would be deduced from the small total amount of Ach extractible from dark fish skin and the usual great difference between the residual Ach of a nervous tissue and the amounts released on its activation (MacIntosh, 1938). However, in spite of this, and with recognition of the efficiency with which Ach reserves may be maintained in active nerves (Brown and Feldberg, 1936), it was believed that extended exposures of fishes to white and to black backgrounds might re-

sult in a measurable difference in the Ach content of the pale and the dark skins. If so, this would yield additional evidence for the view that the melanophore pigment is normally dispersed through the release of Ach. The results of attempts to demonstrate such differences are presented in this paper.

In approaching this problem it might seem at first sight a very simple matter to repeat on the pale skin of a catfish the same kind of test that had been used on the dark skin of this animal. But such is not true, for when the skin of a pale fish is removed preparatory to grinding it, it quickly darkens and thus passes into the opposite color phase from that which it was planned to test. This darkening is due to the stimulation by cutting of the dispersing nerves distributed to the melanophores of the ablated skin. Hence the ordinary removal of pale skin quickly changes it in such a manner as to defeat at the very outset the possibility of the required test. To avoid this difficulty a method must be devised to remove the skin from a pale fish and to keep it in the pale state for the test. To this end several attempts were made in all of which the prime object was to kill the pale skin of the catfish before it had begun to darken and thus to obtain a preparation unquestionably pale in phase.

In the first of these attempts catfishes that had been rendered pale by a sojourn of several days in a white-walled, illuminated tank were taken singly from the tank, decapitated at once with heavy shears, and plunged for fifteen seconds in water at 60°C. By this procedure the skin with its contained melanophores and nerves was quickly killed. It could then be stripped from the fish's body, pulped in a mortar and extracted for testing against standard solutions of Ach with the clam's heart as an indicator; a method based on the findings of Prosser (1940). The critical periods of time from the instant the fishes were lifted from the tank till their skins were killed by the hot water varied from three-quarters of a minute to a minute. The Ach determinations made on a number of fishes whose skins had been prepared in this way were, however, quite diverse nor could any consistency in these differences be discovered. Since, moreover, the skins obtained by these steps did not appear uniformly pale, this technique was abandoned as probably too slow for the object at hand and a second and more rapid technique was tried.

In this second attempt fishes fully blanched were passed directly by hand from the tank water to alcohol chilled with dry ice to about −20°C. At this temperature the fish's skin was not only almost instantly frozen, but in somewhat less than 15 seconds after the fish entered the cold alcohol the fish itself was rigidly solid. Such a fish was then immersed in a large volume of water at 60°C. to insure the death of the skin after which the skin was removed, pulped, and extracted. This second method reduced the critical period during which the skin might darken to about a quarter of a

minute, and skins obtained in this way appeared to the experimenter's eye much more uniformly pale than did those made by the first technique. When, however, the skins prepared by this sudden and intense freezing were subjected to assays for Ach great diversity again reappeared in the results. The failure of both these attempts to obtain uniform Ach determinations from blanched catfish skins led to an abandonment of this type of technique and to the adoption of an entirely different approach to the problem at hand

This new line of attack depended upon the presence on the catfish of a considerable area of permanently pale skin in addition to that which is open to chromatic change. This area of pale skin is on the ventral aspect of the catfish and extends from the region of the opercular folds to that of the pelvic fins. In a fish somewhat over 15 cm. in length the pale area measures about 5 cm. long by some 3 cm. wide. It is white in tint and shows no change when the dorsal coloration of the fish alters under differences in the illuminated surroundings. It is of course not as ideal for this test as an area of blanched skin would be, but it appears to be the best substitute for blanched skin that under the circumstances can be found. When this ventral white skin is removed from a freshly killed fish, it remains permanently white. In making such preparations it is advisable to use catfishes in the dark phase. Under such circumstances two skin preparations can be obtained from each fish, a pale one from its venter and a dark one from its dorsum. Such preparations, in consequence of coming from the same fish, are especially favorable in the comparison of Ach determinations on the two kinds of skin. The detailed steps by which such determinations were made are given in the following description.

Dorsal and ventral areas of skin were removed from two or three fishes and the dark and the pale skins pooled separately. After blotting to remove excess of water, the tissue was placed in tared vessels containing 2 cc. of unbuffered, cold-blooded Ringer, with eserine sulfate 1:5000, and weighed. Normal HCl was added to pH 3 to 4. The skins and fluid were then heated for 3 minutes in a water bath at 90°C. This served the double purpose of aiding the release of bound Ach and greatly facilitating the breaking up of the skin in the grinding process. Additional Ringer was then added to give 1 cc. for each 500 mgm. of tissue. The skins were ground with silica, centrifuged and the clear supernatant fluid decanted for assay. Dilution of the extract with sea water at the time of assay adjusted the pH to within a range favorable to the test preparation.

The extracts of pale and of dark skins were compared with one another and matched against known concentrations of Ach for estimates of their Ach content. The isolated heart of *Venus mercenaria* was used in all tests. The values obtained, as given in table 1, are expressed as the equivalents of the free base in gamma per gram of moist skin.

The results of five separate sets of determinations, each done on a different test heart, are presented in table 1. The estimates for dark skin range from 0.02 to 0.08 gamma of Ach per gram of skin;† those for pale skin from 0.005 to 0.04 gamma per gram. Such variation does not indicate real differences in the Ach content of skins of different groups of fishes, but results rather from the varied response of different hearts to substances in addition to Ach in the extracts. There was evidence of the presence of an inhibitory substance other than Ach in the extracts and differences between hearts, in sensitivity to this substance, doubtless accounts for much of the variation in values for given samples of skin. The relative values of Ach in dark skin to Ach in pale skin in the several sets of experiments are without question significant. They reveal, on the average, over three times as much of this neurohumor in dorsal, melanophore-bearing, dark skin as in ventral, melanophore-free, pale skin.

TABLE 1

FIVE ASSAYS OF CATFISH SKINS, DORSAL DARK AND VENTRAL PALE, FOR ACH EXPRESSED IN FRACTIONS OF A GAMMA PER GRAM OF MOIST SKIN. IN EACH ASSAY TWO OR THREE CATFISHES WERE USED FROM EACH OF WHICH BOTH DARK SKIN AND PALE SKIN WERE TAKEN. ORGANIC INDICATOR: HEART, *Venus mercenaria*

DATES OF TESTS, 1943	ACH, GAMMA PER GRAM OF SKIN		DARK SKIN TIMES PALE
	DARK SKIN	PALE SKIN	
July 8	0.08	0.04−	2+
Nov. 15	0.05	0.015	3.3+
Nov. 16	0.02	0.005−	4+
Nov. 24	0.03	0.01	3
Nov. 26	0.02	0.005	4
Averages	0.04	0.015	3.3

These two types of skin call for careful comparison. Both contain under autonomic control an ample vascular equipment and an abundant supply of mucous glands. These two systems are about equally developed in the dorsal and in the ventral skin. In this respect they are in strong contrast with the autonomically controlled melanophore system which is present in the dorsal skin but absent from the ventral. This distribution of autonomic effectors is fully consistent with the Ach determinations for these two types of skin as recorded in this paper. In the ventral skin where only vascular and glandular components are present the quantity of Ach averages some 0.015 gamma, whereas in the dorsal skin where there is a rich aggregation of melanophores in addition to the vascular and glandular outfits the quantity of Ach reaches on the average 0.04 gamma. Since the excess of Ach in the dorsal skin over that in the ventral, some 0.025 gamma, is thus associated in the dorsal skin with the presence of melanophores, it is natural to conclude that this excess has to do with the system of dark color-cells and that the residue of Ach in the dorsal skin, 0.015 gamma, like that

in the ventral skin is concerned with the other autonomic systems, vasomotor and glandular.

If the conclusion is correct that some 0.025 gamma of Ach per gram of skin is to be relegated to the melanophores, in what way, it may be asked, is this amount associated with these dark color-cells? When the dark skin of a catfish is removed preparatory to testing it for Ach, it is cut off the fish in such a way that it carries with it about a distal millimeter or more of each dispersing nerve-fibre with its numerous branches, chromatic terminals and associated melanophores. The relation of these chromatic terminals to the melanophores is not a simple one. Each melanophore, unlike, for instance, a single muscle-fibre, is innervated by branches from several different chromatic nerve-fibres whose terminals to the number of several scores or more surround the color-cell. Thus a melanophore lies immersed in an aggregation of chromatic terminals instead of being associated with only one efferent terminal as in the case of a muscle-fibre. Where, in this complex, is Ach located and how is it concerned with melanophore activities?

When a catfish darkens it assumes its deep tint as a result of two agencies, intermedine from the pituitary gland and Ach from its nerve supply. Intermedine when injected into a pale catfish can cause it to become fully dark by inducing the pigment in its melanophores to pass from full concentration to full dispersion. Injected Ach, on the other hand, can incite only about half this change in that it can cause the fish to shift its tint from full pale to an intermediate gray, approximately midway between full pale and full dark. If into a catfish rendered intermediate in tone by Ach an appropriate amount of intermedine is injected, the fish will become fully dark. This confirms the view that Ach is a half-darkening activator, so to speak, and intermedine a whole one. Hence after a catfish is hypophysectomized it becomes limited as a result of the loss of its source of intermedine to color changes between pale and intermediate gray. From this standpoint hypophysectomized catfish may be used in the study of the localization of Ach.

If a hypophysectomized catfish with one or more denervated bands cut in its tail is placed in a white-walled, illuminated tank, two changes will occur: The chromatic nerve-fibres and their terminals in the denervated band will degenerate fully in some ten days or so and the whole fish, including its tail and the caudal bands, will become pale. If now this pale fish is transferred to a black-walled, illuminated tank, the whole fish with the exception of its caudal bands will become in the course of a day intermediate gray. The band during and for some time after this change will remain pale. If into such a fish an appropriate quantity of Ach is injected, no change in tint in the fish as a whole will be seen, but the pale bands in its tail will slowly take on a deeper tint till they agree in shade with that of the rest of the tail.

From these observations pertinent conclusions may be drawn. Since the fish devoid of its pituitary gland is without a supply of intermedine such darkening as it is capable of must be due to Ach. As this Ach darkening occurs regularly in innervated areas but never in denervated ones (caudal bands), it is fair to conclude further that the Ach has as its source the nervous portion of the color complex and not the melanophore portion. This view is confirmed by the fact already stated that a pale band, inactive because of the degeneration of its chromatic nerves and terminals, can be darkened by an injection of Ach. Thus Ach introduced from the outside can take the place of the internal chromatic nerves. Hence this evidence appears to rule out the melanophore as a source for Ach and to relegate this material to the nervous portion of the chromatic complex.

This complex in such pieces of skin as have been used in these tests consists of innumerable distal pieces of many-branched chromatic nerve-fibres with a total length of about a millimeter each whose final branches terminate in slightly swollen knobs, the chromatic terminals. These terminals as already stated are in close proximity to the melanophores. Is Ach a product of this nervous mechanism as a whole or of one of its parts, the branches of the dispersing chromatic nerve-fibres or the terminals? No conclusive answer can be given to this question, but some evidence concerning it can be found in the exact relation of denervated dark areas to the locations of the cuts in the nerves whereby these dark areas are induced. Such conditions are well illustrated in certain relations shown by the ophthalmic nerve in the catfish. This nerve and its branches pass anteriorly over the dorsal wall of the orbital space in this fish to emerge dorsally through a thin layer of bone and spread out over the under surface of the skin in their distribution to the anterior dorsal region of the fish's head. For the test at hand these branches may be cut at the anterior limits of the orbit where they pass out of this cavity and through the bone to gain the inner surface of the skin. Shortly after these nerves have been cut the area of skin thus denervated on the fish's head will be easily distinguished by its dark tint and it is a noteworthy fact that this denervated, dark area does not begin where the nerve branches enter the skin but appears first several millimeters anterior to this region. In other words the chromatic nerve-fibres pass anteriorly through the skin on the fish's head some distance before they reach the darkened area. This relation indicates that the chromatic nerve-fibres of themselves are probably in no direct way concerned with the darkening of the skin. It is only after the region of their terminals has been reached that the darkening of the skin is to be seen. Hence this evidence indicates that the chromatic nerve-fibres in themselves have no direct concern with the darkening response, but that this response must be induced by the distal ends of these fibres, in other words, by their terminals. Although this evidence as already intimated cannot be said to be fully conclusive, it is not

without weight. It certainly suggests a consistent picture of the relations of the so-called chromatic terminals to the darkening process.

Assuming the correctness of this conclusion these terminals must be the parts immediately concerned with the 0.025 gamma of Ach which is proper to the melanophore apparatus in a single gram of dark skin. Whether this extremely small amount of Ach is all carried in the fish's lymph and other tissue fluids directly next the dark color-cells or whether it is in part in these fluids and in part in the chromatic terminals cannot be stated. In our opinion Ach is formed in the terminals where it is for the most part stored to be liberated in minute amounts through the action of chromatic-nerve impulses in the normal darkening of the fish or released as a whole from the terminals in the process of artificial extraction. We hold this opinion notwithstanding the fact that the total amount of Ach as measured by us is in itself extremely small and that the added reduction implied in the last statement places the functional amounts of Ach in the darkening of fishes at almost inconceivably small figures.

Summary.—1. The dark dorsal skin of the catfish Ameiurus containing three autonomically controlled systems (blood-vascular, mucous glands and melanophores) yielded from 0.02 to 0.08 gamma (average, 0.04) of Ach per gram of moist skin.

2. The pale ventral skin of the catfish containing only two autonomically controlled systems (blood-vascular and mucous glands) yielded from 0.005 to 0.04 gamma (average, 0.015) of Ach per gram of moist skin.

3. The difference between the averages of these two yields, 0.025 gamma of Ach, appears to be the amount of this neurohumor concerned with the activity of the melanophores.

4. This Ach is not produced by the melanophores. It probably does not originate in the dispersing chromatic nerve-fibres but rather in their terminals where it is very likely stored to be used from time to time as an agent to excite the dispersion of melanophore pigment.

5. Admitting this interpretation of melanophore activation by Ach it would appear that this operation calls for an amount of this neurohumor almost infinitesimally small.

* The expenses of this investigation were met by a grant from the Permanent Science Fund of the American Academy of Arts and Sciences. We wish to express here our obligations to the officers of this Fund for their generous help.

† It is to be noted that the Ach determinations already quoted from Chang, Hsieh and Lu (1939) and from Parker (1940), namely, 0.077 gamma and 0.078 gamma, respectively, per gram of dark skin, fall within the range of determinations for dark skin given in table 1.

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INADEQUACIES IN PRESENT KNOWLEDGE OF THE RELATION BETWEEN PHOTOSYNTHESIS AND THE O^{18} CONTENT OF ATMOSPHERIC OXYGEN

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Communicated November 7, 1944

Various lines or evidence strongly indicate that the oxygen of the atmosphere has been formed mainly by photosynthesis. If this conclusion is assumed to be correct, it follows that any specific relation between the substrates and products of photosynthesis that can be demonstrated in the laboratory should also be observable on a vastly larger scale in nature provided other interfering factors are not operative. One such specific relation with which we shall be concerned is that between the O^{18} content of water and of photosynthetic oxygen. The O^{18} content of the oxygen produced by the green alga *Chlorella pyrenoidosa* and by two land plants, sunflower and coleus, has been shown to be determined by the isotope content of the water and to be independent of that of the available carbon dioxide. Consequently, one might expect that in nature the O^{18} content of the oxygen of the atmosphere would be equal to that of the water in the oceans, since it is estimated that about four-fifths of the total photosynthesis occurs there and would be different from that of naturally occurring carbon dioxide and carbonates. Actually just the reverse is true; the O^{18} content of atmospheric oxygen is closer to that of carbonates than to that of ocean water, the O^{18} content being higher in the carbonates and atmospheric oxygen than in water.

A careful examination of the pertinent biological and physical evidence has led us to the conclusion that this apparent inconsistency does not constitute valid evidence against the photosynthetic theory of the origin of

atmospheric oxygen. It appears probable that an eventual explanation will be found in terms of isotope exchange reactions between molecular oxygen and water or other oxygen-containing compounds. However, the available data on isotope equilibria in water-oxygen systems cannot explain the observed relations. Therefore, a further study of such systems is desirable. Also additional information on the ultimate origin of oxygen produced by marine plants is needed to verify and extend the results obtained with fresh water and land plants.

A more detailed discussion of the points mentioned above is given in the following sections.

Geochemical Evidence for the Photosynthetic Origin of Atmospheric Oxygen.—In support of the assumption that atmospheric oxygen has been and is now being produced according to the well-known equation of photosynthesis

$$n\,CO_2 + n\,H_2O = n\,(CH_2O) + n\,O_2 \qquad (1)$$

we shall mention only two types of geochemical evidence. First, the estimated quantity of reduced carbon of biological origin on the earth's surface in the form of coal, oil, peat, humus, etc., corresponds approximately to the quantity of oxygen in the atmosphere after a correction has been made for oxygen removal by the weathering (oxidation) of igneous rocks and other similar reactions.[1] Second, the present rate of photosynthesis is sufficient to produce the quantity of oxygen in the atmosphere in a few thousand years, thereafter maintaining the atmosphere at its present oxygen level.[2] From this latter fact it may be concluded that whatever the origin of the atmosphere, the oxygen now present there must have been formed mainly by photosynthesis.

Basic Facts Concerning Oxygen Isotopes and Isotope Exchange Reactions.—Before discussing the experiments on the origin of oxygen in photosynthesis and the distribution of O^{18} in nature, it will be helpful to review briefly some basic facts concerning oxygen isotopes and isotope exchange reactions.

Natural oxygen contains three isotopes with mass numbers 16, 17 and 18, and percentage abundances of 99.76, 0.041 and 0.20, respectively.[3,4] In "enriched" samples of oxygen the increase in weight is usually credited to the O^{18} isotope entirely since the O^{17} contribution is relatively small due to its low abundance. In this paper the term "tracer oxygen" will be reserved for oxygen artificially enriched with O^{18}. The term "natural" oxygen will be understood to mean oxygen with the above mentioned isotope composition.

If two oxygen-containing molecules such as water and carbon dioxide are allowed to react until isotope equilibrium is reached, they do not in general contain the same proportions of isotopes. The relative abundance of isotopes in the two molecules may be expressed by a quantity known as the "enrichment factor," defined by the ratio $(N_1/N_2)/(n_1/n_2)$ where N_1 and

N_2 are the numbers of each isotope in one compound and n_1 and n_2 are the corresponding numbers in the other compound. Enrichment factors for several reactions which are involved in the present discussion are given in table 1, along with the corresponding equilibrium constants. The values have been calculated[5] on the basis of theoretical considerations; experimental verification[6] is available only for reaction (3).

Examination of table 1 shows that the enrichment factors for all the reactions here considered are larger than unity; this means that there is a higher proportion of oxygen O^{18} in the carbon dioxide or oxygen than in the water in isotopic equilibrium with it. Reactions (3) and (5) are of greatest importance for the present discussion. In the system CO_2 (g)–H_2O(1), (reaction (3)), at 25°C., the O^{18} enrichment in the gaseous carbon dioxide is such that water prepared from it is denser than the water in equilibrium with it by some ten gamma units.* The kinetics of the reaction are such that equilibrium can be attained in a relatively short time under suitable conditions. The reaction is slowest but still measurable in alkaline solutions (pH = 10) in which the carbon dioxide is mainly in the form of the carbonate ion. As the pH is lowered into the acid range (pH $\lesssim$ 7), the rate increases rapidly.[7] In the equilibrium system O_2(g)–H_2O(1), (reaction (5)), at 25°, water prepared from the oxygen has a density excess of about 1 gamma unit over the water. Nothing appears to be known about the kinetics of this reaction, but the rate is presumably very slow.

TABLE 1

ENRICHMENT FACTORS AND EQUILIBRIUM CONSTANTS OF SOME OXYGEN EXCHANGE REACTIONS (AFTER UREY AND GREIFF)

REACTION	EQUILIBRIUM CONSTANT 0°	EQUILIBRIUM CONSTANT 25°	ENRICHMENT FACTOR 0°	ENRICHMENT FACTOR 25°
(2) $CO_2^{16}(g) + H_2O^{18}(g) = CO_2^{18}(g) + H_2O^{16}(g)$	1.128	1.110	1.064	1.054
(3) $CO_2^{16}(g) + H_2O^{18}(l) = CO_2^{18}(g) + H_2O^{16}(l)$	1.097	1.080	1.047	1.039
(4) $O_2^{16}(g) + H_2O^{18}(g) = O_2^{18}(g) + H_2O^{16}(g)$	1.048	1.041	1.024	1.020
(5) $O_2^{16}(g) + H_2O^{18}(l) = O_2^{18}(g) + H_2O^{16}(l)$	1.020	1.012	1.010	1.006

For the study of photosynthesis it is also important to know the relative O^{18} contents of dissolved and gaseous carbon dioxide in equilibrium. At present there is no direct experimental evidence on this point. However, it may be inferred that carbon dioxide in the gaseous and dissolved states has about the same O^{18} content from the fact that calcareous rocks of marine origin have essentially the same isotope content as gaseous carbon dioxide in equilibrium with water.[8] Obviously direct experimental comparison of the O^{18} contents of dissolved and gaseous carbon dioxide would be most desirable; this could be done by a study of carbonate precipitated from equilibrated carbon dioxide–water systems.

An additional observation of importance must also be mentioned here. Water containing O^{18} has a slightly lower vapor pressure than water containing O^{16}. Therefore, during the evaporation of ordinary water, O^{18} tends to concentrate in the residual liquor.[9]

For determining the O^{18} content of a compound, two general methods are available; one involves measuring the density of water prepared from the oxygen and the other involves the direct determination of the relative numbers of oxygen atoms of different weights by means of a mass spectrometer. The former method is more sensitive, while the latter has the advantage of requiring only very small oxygen samples.

For measuring very small density differences, the so-called "submerged float" method provides a very satisfactory technique.† In this method a small glass or quartz bubble is immersed in the liquid, the density of which is to be measured. The temperature at which the float neither sinks nor rises is determined to about 0.0002°C. and is compared with the corresponding temperature for a solution of known density. From the temperature difference and a knowledge of the rate of density change with temperature, the relative density and therefore the relative O^{18} content of the sample can be calculated. With a temperature control of 0.0002°C., a density difference of about 0.5 parts in ten billion or .05 gamma unit should be detectable. Such sensitivities are far greater than warranted by the purification procedures employed which, in general, limit reproducibility of measurements to a few tenths of a gamma unit. In the experiments to which we shall refer in the following two sections, a density difference of 1 gamma unit can be regarded as significant. Details of the method are given in references cited below.**

The sensitivity of the mass spectrometer method of estimating O^{18} varies somewhat with the design of the instrument, being 0.5–1.0 gamma unit with the best models and ranging up to 5 or 10 gamma units density difference with the less sensitive models. This method, therefore, is not suitable for the study of the very small differences in O^{18} content such as exist, for example, in equilibrium systems of water and carbon dioxide, unless a very good instrument is available.

With the material presented above in mind, we can now proceed to consider the experiments on the origin of oxygen in photosynthesis and the known facts concerning the O^{18} distribution in nature.

Studies on the Rôle of Water as the Source of Photosynthetic Oxygen.—Since the substrates of photosynthesis are water and carbon dioxide, it is obvious that the evolved oxygen must be derived ultimately from one or both of these compounds. Various theoretical considerations, which need not be reviewed here, lead to the conclusion that the oxygen of photosynthesis probably comes from water and not from carbon dioxide.[10] The first experimental study of this hypothesis was undertaken in 1941 making

use of tracer oxygen.[11] Because a knowledge of the ultimate source of photosynthetic oxygen is of great importance in interpreting data on the distribution of oxygen isotopes in nature, the evidence bearing upon this hypothesis will be discussed in some detail.

Let us first examine the logic underlying experiments on the origin of photosynthetic oxygen. To demonstrate that oxygen originates from water, for example, it is necessary to "label" the oxygen in the water and show that it is transformed into molecular oxygen by the photosynthetic process. It is also necessary to show that when oxygen in carbon dioxide is labelled, no tracer oxygen appears as molecular oxygen in the gas phase. Since all photosynthetic products are derived from water or carbon dioxide, this demonstration would be sufficient to establish water as the ultimate source of oxygen. In carrying out such an experiment, one serious complication arises. If either the water or the carbon dioxide is labeled with tracer oxygen, the isotope exchange reaction (3) will immediately begin to redistribute the O^{18} and eventually isotope equilibrium will again be reached. Because of this exchange, the initial relatively large difference in isotope contents of water and carbon dioxide will be effective in labeling these molecules only when the rate of redistribution of O^{18} is small in comparison to the rate of photosynthesis. If the rate of exchange is relatively great, isotope equilibrium will be approached before the substrate molecules have had time to enter into the photosynthetic reaction and the isotope contents of the carbon dioxide and the evolved oxygen will necessarily approximate that of the initial water because the latter is present in vastly greater amount. It is therefore of great importance in experiments of this sort to provide proof that the rate of isotope exchange is too slow to invalidate the results.

In the experiments of Ruben, *et al.*, cell suspensions of *Chlorella pyrenoidosa* were allowed to photosynthesize in a carbonate-bicarbonate buffer solution having a pH of about 10. In some experiments the water was labelled with tracer oxygen, in others the carbonate was labeled. In both cases, the O^{18} content of the evolved oxygen was equal to that of the water within the limits of accuracy of the mass spectrometer measurements and differed widely from that of the carbon dioxide. As was to be expected from the kinetic studies already cited, a slow exchange of O^{18} was observed to occur between the carbon dioxide and the water. The overall rate of this exchange under the experimental conditions was slow enough to be neglected in comparison with the rate of photosynthesis. It was therefore concluded that the oxygen originated solely from water.

Such a conclusion is reasonable, but by no means certain since it depends on the unproven assumption that the isotope exchange is no more rapid inside the cells, and especially in the chloroplasts where photosynthesis actually occurs, than in the outside medium where it was measured.

It is quite possible and, indeed, even probable that this assumption is incorrect since the rate of isotope exchange increases rapidly with increasing hydrogen ion concentration, and it is known that many plant cells have an internal vacuolar pH $\leqq$ 6, which is maintained more or less constant despite wide variations of pH in the external medium.[12] It can be calculated that at pH 6, the randomization of O^{18} is rapid enough to invalidate the conclusion that carbon dioxide is not a source of oxygen.

In view of the existing uncertainty as to the internal pH of the *Chlorella* cell and the consequent uncertainty as to the rate of the randomization of O^{18} at the site of photosynthesis, these experiments do not provide proof of the rôle of water as the precursor of oxygen.

Recently, new and apparently unambiguous experiments[13,14] on the origin of photosynthetic oxygen have been performed which support the previous conclusion that oxygen arises wholly from water. The above-mentioned difficulties resulting from randomization of O^{18} were avoided by making use of the small difference between the O^{18} contents of water and carbon dioxide in equilibrium to trace the origin of the photosynthetic oxygen. This was made possible by recourse to the very sensitive submerged float method to analyze for O^{18}. In these experiments, *Chlorella* suspensions were allowed to photosynthesize in ordinary water equilibrated with carbon dioxide by means of the enzyme carbonic anhydrase from beef blood. The O^{18} content of the evolved oxygen was found to be nearly the same as that of the water and significantly lower than that of the carbon dioxide. Experiments with land plants (sunflower and coleus) yielded similar results.

It seems reasonable to conclude that the rôle of water as the precursor of oxygen has been established in the case of one fresh water alga, *Chlorella pyrenoidosa*, and two land plants, sunflower and coleus. However, it must be remembered that most photosynthesis occurs in the oceans and hence marine organisms are much more important as large-scale producers of oxygen than are fresh water algae or land plants. It is therefore desirable that the above experimental results should be verified and extended by studies on a number of typical marine algae. There is no reason to believe that such studies will show any significant differences from those already reported. However, pending their completion, any general statement concerning the origin of photosynthetic oxygen can be accepted only with some reservation. Also there exists the possibility that during photosynthesis isotope equilibrium is established between the evolved oxygen and water. No evidence for such equilibration was obtained in the experiments of Ruben, *et al.* However, Dole and Jenks found that the O^{18} content of photosynthetic oxygen is approximately 1 gamma unit higher than that of the water. This suggests that equilibration may have occurred in these experiments. Obviously, the possibility that Chlorella catalyzes the ex-

change equilibrium between gaseous oxygen and water should be reinvestigated.

The Distribution of O^{18} in Nature.—The available information on the distribution of O^{18} in nature may be summarized as follows.[15]

(1) Oxygen in fresh water has the same O^{18} content as that in rocks and ores not containing carbonates.

(2) Sea water has a slightly higher O^{18} content than fresh water. The work of a number of investigators shows a density excess of 1.3–2.3 gamma units.[15,16] The increase in density is in good agreement with the value predicted from the experimental results obtained in isotopic fractionation of water during evaporation.

(3) Water prepared from the oxygen of carbonate rocks is denser than fresh water by some 8 gamma units.[8] This density excess is close to the value predicted on the basis that the carbonate is deposited from carbon dioxide in the sea water which is in equilibrium with atmospheric carbon dioxide and consequently enhanced in O^{18} according to reaction (3).

(4) Water prepared from atmospheric oxygen shows a density excess over fresh water of 6.6 gamma units.[8] It is here that the discrepancy arises between the physical data and the demonstration that photosynthetic organisms evolve oxygen with an O^{18} content very near that of water. 2 gamma units can be accounted for as due to the effect of evaporation discussed in (2) above. This leaves a 4.6 gamma unit discrepancy. On the basis of reaction (5), assuming a temperature intermediate between 0° and 25°C. and the attainment of equilibrium, one can predict a further concentration of O^{18} in the gaseous oxygen corresponding to a density excess of about 1 gamma unit. This effect has been observed in the experiments of Dole and Jenks.[14] The residual 3.6 gamma units, sometimes known as the "Dole effect," remain to be explained.

As mentioned previously, the suggestion[15,17] that carbon dioxide enriched in O^{18} contributes the necessary heavy oxygen during photosynthesis is invalidated by all the experimental evidence so far obtained. There remain the following possibilities:

(*a*) The enrichment factor calculated for reaction (5) is in error. This is extremely doubtful since the physical constants required for the calculation of this equilibrium are accurately known. The theoretical values for this reaction should in any event be as reliable as those for reaction (3) which has been experimentally verified.

(*b*) Reaction (4), which has a more favorable enrichment factor than reaction (5), may be contributing to the O^{18} enhancement of atmospheric oxygen. However, at or near the surface of the earth where the oxygen studied was collected, reaction (5) must dictate the equilibrium finally reached, because the O^{18} content of the water involved would be determined by the large amount of liquid water in comparison with the water vapor.

As we have already remarked, reaction (5) fails by a large margin to account for the Dole effect.

(*c*) As previously mentioned, it is possible, though unlikely, that Chlorella and the two land plants so far studied are not typical of photosynthetic plants, particularly marine algae, with respect to the origin of their oxygen. This possibility must be investigated.

Although an ultimate explanation of the Dole effect may be found to depend upon either a biochemical or a physical effect, the latter possibility seems to us much more probable. In any event the existing inconsistency between the experimental evidence on the source of photosynthetic oxygen and the distribution of oxygen in nature cannot be regarded as valid evidence against the biological theory of the origin of atmospheric oxygen.

The authors wish to express their appreciation to Professors R. T. Birge, D. R. Hoagland and M. Dole, H. C. Urey and J. Franck for numerous valuable suggestions. We are also indebted to Professor Dole for permission to quote the results of unpublished work by him and G. Jenks.

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PRODUCTION OF STAPHYLOCOCCUS STRAINS RESISTANT TO VARIOUS CONCENTRATIONS OF PENICILLIN*

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Communicated December 5, 1944

It is a well-established fact that strains of bacteria resistant to various sulfa drugs, as well as strains resistant to penicillin, may readily be obtained by growing bacteria on media containing increasingly higher concentrations of the respective chemicals. The purpose of this study was to make a quantitative survey of the origin of resistant bacteria, and to clarify the genetic aspect of the mechanism through which resistance is formed. A preliminary report summarizing the results obtained is given below.

Material and Method.—A strain of *Staphylococcus aureus* obtained from the Northern Regional Research Laboratory, Peoria, Illinois, carrying the N.R.R.L. number 313, was used in these experiments. This particular strain is employed by several laboratories for assaying penicillin. Before the experiment was started, a broth culture was prepared with bacteria from a single colony, and from this broth culture three agar slants were inoculated. These three stock cultures were kept in a refrigerator and served daily as the source of inoculum for all experiments. In a long series of experiments conducted over a considerable period of time, this procedure yields material that should be genetically more uniform than if the stock were maintained by consecutive transfers.

Penicillin was taken from a lot of sodium salt of penicillin prepared by E. R. Squibb and Sons, New York, which was packed in ampules containing 25,000 Oxford units each. The material of one ampule was dissolved in 10 cc. of phosphate buffer of pH 6, and kept in the refrigerator as a stock solution containing 2500 Oxford units of penicillin per cc. From this, other stock solutions containing 250 and 25 units per cc. were prepared under sterile conditions. Assays made at intervals indicated that the potency of penicillin in the stock solutions was not affected by storage.

The resistance of the bacteria to penicillin was determined by mixing them with an agar-nutrient medium to which the penicillin solution had been added, and plating the mixture in a Petri dish. Precautions were taken not to have the agar warmer than 45 degrees Centigrade.

In order to have a check of the potency of the penicillin, an assay was made for every experiment by diluting the solution used in that experiment to one Oxford unit per cc. and making a standard Oxford cup test. In this way any appreciable decrease in the potency of the penicillin solution would have been detected.

Reaction of Staphylococcus to Penicillin.—The strain of bacteria used in

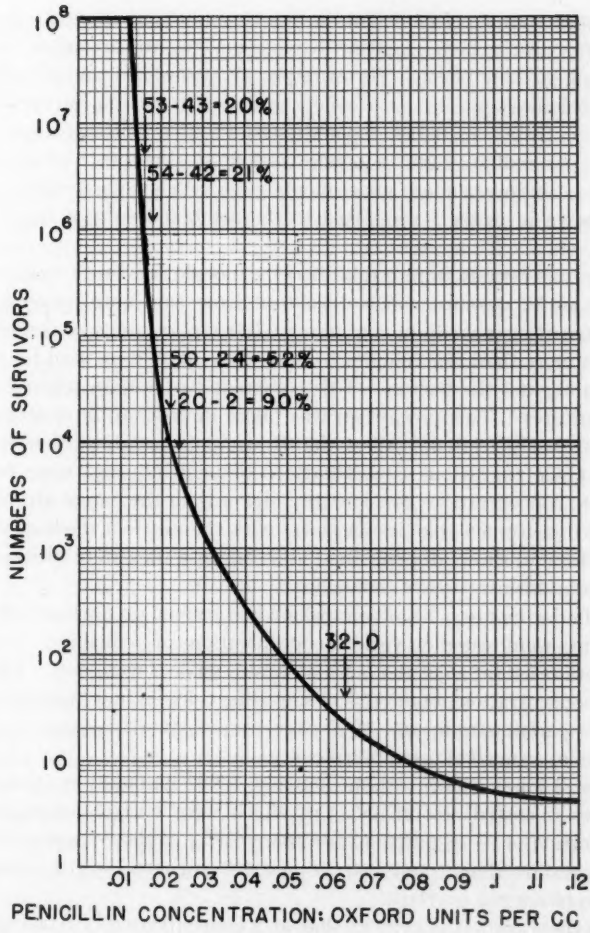

FIGURE 1

Numbers of surviving *Staphylococcus aureus* after plating on nutrient agar containing various concentrations of penicillin. Arrows indicate concentrations at which resistance to penicillin of surviving colonies was tested, and numbers above the arrows show the total number of colonies tested, the number of normal overlaps and the percentage of resistant colonies.

these experiments was affected by various concentrations of penicillin in a manner shown graphically in figure 1. No reduction in colony counts is apparent when the bacteria are plated onto nutrient-agar medium containing

weak concentrations of penicillin, until a threshhold of about 0.012 Oxford units per cc. is reached. A slight increase in concentration after this point produces a striking effect. For example, at a concentration of 0.014 only 10% of colonies appear; at a concentration of 0.016 the survival is 1%; at a concentration of 0.018, 1 per thousand; at a concentration of 0.05, about 1 per million; and a concentration of about 0.15 eliminates all bacteria. Six independent experiments gave very similar results, and the curve shown in figure 1 was fitted to the combined data of these experiments.

Thirty-two strains were established by inoculating broth with bacteria taken from 32 colonies which developed from surviving bacteria plated onto nutrient-agar medium containing 0.064 Oxford units of penicillin per cc. Not more than two colonies from any one plate were used for starting new strains, and the bacteria of the different plates were taken from different cultures. This precaution was taken in order to have strains that were not closely related. All of the 32 strains so established were tested for resistance to various concentrations of penicillin and were found to withstand concentrations considerably higher than the parent strain. The bacteria that formed colonies on plates with the nutrient medium containing 0.064 units per cc. of penicillin did so because they were resistant to at least that concentration of penicillin.

In a similar manner, 20 colonies which had formed on medium containing 0.024 units were tested on various concentrations of penicillin; 18 of them were more resistant than the original strain, while 2 were not. Therefore, at that concentration about 90% of surviving colonies had higher resistance than the parent strain, and 10% were normal overlaps—that is, chance survivors in a survival probability curve.

At a still lower concentration—namely 0.022 units per cc.—there were 50 survivors tested, 24 (or 48%) of which were normal overlaps. At a concentration of 0.018 units, 42 (or about 79%) of the 54 survivors tested were normal overlaps; and at 0.016 units, 43 (or 80%) of the 53 survivors tested were normal overlaps.

From these tests it is evident that at a concentration of 0.064 units per cc. of penicillin all tested survivors had higher resistance than the parent strain, while even at lower concentrations a considerable proportion of survivors were resistant. In figure 1 the arrows indicate concentrations from which survivors were tested, and the numbers above the arrows show the total number of tests, the number of normal overlaps and the percentage of resistant colonies.

Is the Resistance Inherited?—The evidence that has been accumulated suggests an affirmative answer to this question. A resistant strain isolated from the medium containing 0.064 units of penicillin was kept in the refrigerator on an agar slant for three months without any change in the de-

gree of resistance. Ten strains isolated in a similar manner were passed through 20 broth transfers, and tests for resistance were made at the end of that period as well as several times during the process. No change in the degree of resistance was observed. These strains, which were isolated after one passage in penicillin, acquired permanent resistance.

Origin of Resistant Bacteria.—Two alternate mechanisms can be visualized as responsible for the origin of bacteria resistant to certain concentrations of penicillin: (1) Resistance is an acquired characteristic, which develops through interaction between bacteria and penicillin when the two are in contact with each other. (2) Resistance is an inherited characteristic, which originates through mutation and whose origin is independent of penicillin treatment; resistant mutants occur at random, in a small fraction of a population, and, since a certain concentration of penicillin eliminates all non-resistant individuals, the resistant ones are selected out from the population by the treatment.

Which of these two mechanisms is responsible for the origin of resistance can be determined with the aid of a modification of the method developed by Luria and Delbrück[1] in their study of changes in bacteria from bacteriophage-sensitivity to bacteriophage-resistance. In the majority of experiments reported in this paper, bacteria were in contact with penicillin only during the time when the test for resistance was being carried on. Otherwise they were grown in the broth medium free of penicillin. If the resistance is induced through interaction between bacteria and penicillin when they are in contact with each other, it would be expected that approximately similar numbers of resistant bacteria would be obtained when samples containing similar numbers of bacteria are plated onto nutrient agar containing a certain concentration of penicillin, irrespective of the origin of these samples. The situation would be quite different in the event that the origin of resistance is mutational. In such case, one would expect to obtain similar numbers of resistant colonies only in samples taken from the same culture. If, however, each of the samples came from a separate culture, and mutations occur at random, then one would expect to obtain a large number of resistant colonies from cultures in which mutation happened to occur early in the growth of the culture and a small number of resistant colonies from cultures in which mutation happened to occur late, provided resistant bacteria grow more or less like the normal ones. If resistance originates by mutation, then, the variation in number of resistant bacteria between samples taken from separate cultures should be much greater than between samples taken from the same culture.

A critical experiment to distinguish between these two possibilities was planned as follows: A saturated broth culture of bacteria was diluted to 10^{-6}, so that the broth contained about 300 bacteria per cc. 0.3 cc. of this was placed in each of 30 small test tubes. One large test tube contain-

ing about 15 cc. of broth was inoculated with another sample of 0.3 cc. from the same dilution. All 31 test tubes were incubated at 37°C. for 18 hours, and precautions were taken to prevent evaporation from the tubes containing the small cultures. After 18 hours of incubation, 0.7 cc. of broth was added to each of these 30 small tubes, in order to reduce the error when the contents of each tube were taken out and plated. The number of bacteria was determined by sampling 10 of the small tubes, taking 0.05 cc. of material from each and assaying it on nutrient agar after making proper

TABLE 1

NUMBER OF BACTERIA RESISTANT TO CONCENTRATION OF 0.064 OXFORD UNITS OF PENICILLIN PER CC. OF AGAR MEDIUM IN SAMPLES TAKEN FROM A SERIES OF INDEPENDENT CULTURES AND SIMILAR SAMPLES TAKEN FROM A SINGLE CULTURE WHICH ASSAYED 2.3×10^8 BACTERIA PER CC.

SAMPLES FROM INDEPENDENT CULTURES					SAMPLES FROM A SINGLE CULTURE	
CULTURE NO.	NO. OF BACTERIA PER CC.	NO. OF RESISTANT BACTERIA	CULTURE NO.	NO. OF RESISTANT BACTERIA	SAMPLE NO.	NO. OF RESISTANT BACTERIA
1	1.83×10^8	33	11	196	1	27
2	1.79	18	12	66	2	35
3	1.82	839	13	28	3	34
4	1.79	47	14	17	4	32
5	2.02	13	15	27	5	33
6	2.05	126	16	37	6	27
7	1.76	48	17	126	7	25
8	1.85	80	18	33	8	28
9	2.06	9	19	12	9	34
10	2.02	71	20	44	10	38
			21	28	11	25
			22	67	12	29
			23	730	13	31
			24	168	14	38
			25	44	15	31
			26	50	16	23
			27	583	17	16
			28	23	18	21
			29	17	19	30
			30	24	20	21
Average	1.9×10^8	128.4	..	116	..	28.9
Variance	1.35	57255	..	35399	..	39.8
χ^2	7.082	4459	..	6103	..	22.7
P	0.6	.....	..	...	..	0.3

serial dilutions. The number of bacteria in the large culture was determined by a similar assay. The material from the 30 small cultures (containing 0.3 cc. of saturated bacteria to which 0.7 cc. of broth had been added) was plated onto Petri dishes with nutrient agar containing 0.064 Oxford units of penicillin per cc. At the same time, twenty 0.3-cc. samples containing saturated bacteria from the large culture were plated in agar with 0.064 units of penicillin per cc.

Results of this experiment are given in table 1. It may be seen that the growth of bacteria in the different small cultures was fairly uniform, the average titre in ten cultures being 1.9×10^8 of individuals per cc.; and that this was fairly close to the titre of 2.3×10^8 per cc. reached in the large culture. That is, samples taken from the individual cultures and from the large culture contained approximately similar numbers of bacteria. It is evident from the table that the variation between the numbers of resistant bacteria on plates from samples taken from a single culture is small, the extremes being 16 and 38, the variance slightly larger than the average, and the probability that this variation is due to chance 30 per one hundred trials. On the other hand, the variation in number of resistant bacteria among samples taken from independent cultures is considerable, with extremes of 9 and 839 in cultures number 1 to number 10, and 12 and 730 in cultures numbers 11 to 20, with a variance greater than 200 times the average, and an insignificant probability that such a distribution may be due to sampling.

The results of this experiment, therefore, favor the assumption that resistance to certain concentrations of penicillin originates through mutation and that resistant bacteria may be found in any large population. The proportion of resistant bacteria depends on the mutation rate. The experiment was repeated three times, and similar results were obtained.

Another experiment furnished supporting evidence for the conclusion that resistance does not originate as a result of contact between bacteria and penicillin. In a study of the action of penicillin on Staphylococcus, it was found that penicillin affects principally dividing bacteria, while nondividing bacteria can be kept for a considerable length of time in the penicillin-containing medium. To a saturated broth culture, containing 3.7×10^8 bacteria per cc., a sufficient amount of penicillin was added to make the concentration in the medium 25 Oxford units per cc. The culture was kept at 37°C., and after five days an assay showed that it contained 2.3×10^8 living bacteria per cc., while the assay for penicillin indicated that the concentration was not appreciably changed. These bacteria, exposed to 25 units of penicillin for five days, were washed to remove penicillin from the medium and tested for their resistance to various concentrations of this chemical. They were found to be no more resistant to penicillin than bacteria of the original strain. This shows that contact with penicillin does not make resting bacteria resistant.

Degree of Resistance.—In the strain of Staphylococcus used in these experiments, there is about one bacterium per 2×10^8 that survives a concentration of penicillin of 0.125 units per cc. At a concentration of 0.15 units per cc., there are no survivors. However, in strains established from survivors on an 0.064 concentration, there were a few individuals resistant to 0.15 units; in strains developed from survivors on an 0.125 concentration,

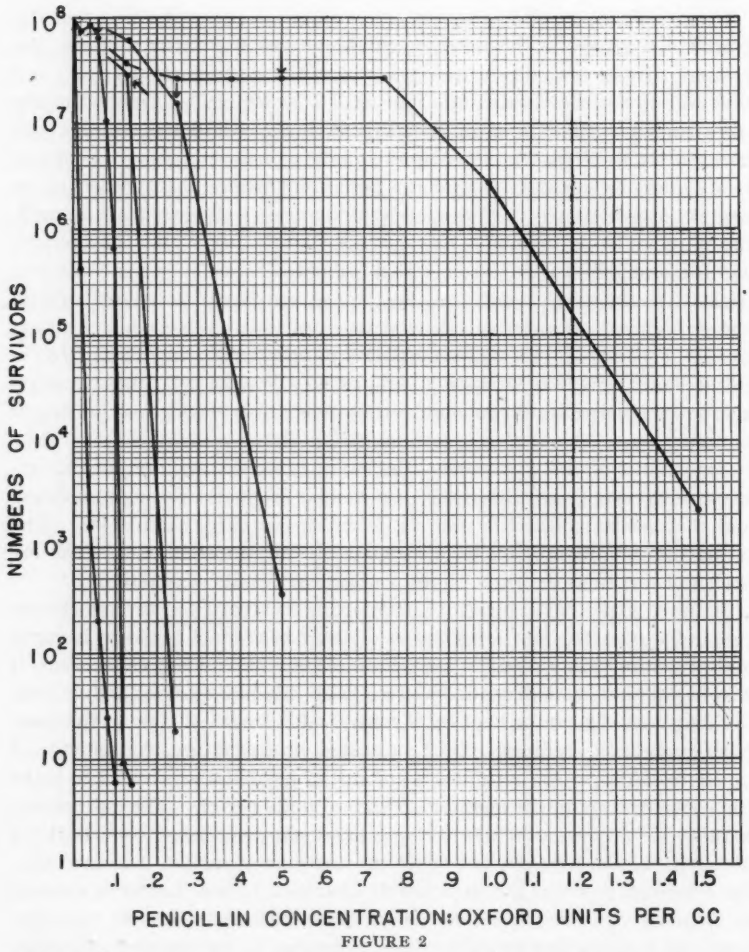

FIGURE 2

Numbers of survivors on various concentrations of penicillin, for the stock strain of *Staphylococcus aureus* and for four resistant strains developed through repeated selection.

there were individuals resistant to 0.25 units; strains from these latter survivors had individuals resistant to 0.5 units; strains from these contained individuals resistant to 4 units; and from these a strain was isolated that was not affected by a concentration of 250 units of penicillin per cc. of the agar medium.

The curves in figure 2 show the survival numbers, with various concentrations of penicillin, for bacteria of the stock strain and of four strains developed by the process of selection described in the previous paragraph. It is evident that the building up of resistance is more rapid with each selection step. That is, a concentration of 0.15 units is sufficient to eliminate all bacteria of the original strain, while to eliminate all bacteria of the first-step resistant strain a concentration of about 0.2 units is required, for the second-step resistant strain about 0.4 units, for the third-step about 1.0 units, and for the fourth-step about 7 units. The fifth-step strain was for all practical purposes completely resistant to penicillin.

As a check against contamination, a bacteriophage strain was isolated which lyses the Staphylococcus strain used in these experiments. This phage lysed also the completely resistant strain mentioned above, and all other resistant strains with which it was tested, indicating that these strains were derived from the original one rather than contaminants.

Discussion.—The evidence reported in this paper makes it probable that resistance of Staphylococcus to certain concentrations of penicillin is not induced by the action of penicillin on bacteria, but arises independently by mutation.

In any large population of bacteria of the strain of Staphylococcus used in these experiments there are some individuals resistant to certain low concentrations of penicillin. If this population is exposed to the action of such concentrations of penicillin, non-resistant individuals are eliminated while the resistant survive. Thus penicillin acts as a selective agent which suppresses non-resistant bacteria.

It is clear from the data that resistance is a complex characteristic, and that it must involve a number of mutations; if it is assumed that genes are responsible for these mutations, a number of genic changes must be involved. Such a situation is not unusual. A close parallel was described by Demerec and Fano[2] in the case of strain B of *Escherichia coli*, where about 20 distinguishable mutant types showing resistance to one or more of the seven phages were detected. Since it would be possible to isolate many more phages affecting the B strain of coli, it is evident that the actual number of mutants affecting resistance is considerably larger than the number detected.

It has been shown that degree of resistance can be increased by selection, and that the building up of resistance is more rapid with each selection step (Fig. 2). This can readily be explained by the mutation hypothesis. It is assumed that there are a number of genes that affect resistance to penicillin, and if any one of them mutates, the individual in which such mutation occurs acquires resistance to a certain concentration of penicillin. Mutants may differ in degree of resistance, but the resistance of individuals in which only one gene has mutated (single mutants) is never very high.

In a single-mutant strain, mutations may occur in other genes for resistance; and when two mutant genes are together in one individual (double mutants) their effect is cumulative. Moreover, it happens that the resistance of a double mutant is higher than the sum of resistances of two single mutants. If a third gene for resistance, a fourth, etc., mutate in the same line, the combined effect of all these mutations is a high degree of resistance or complete resistance.

Acknowledgments.—Acknowledgment is made of the very efficient assistance rendered by Miss Marion N. Crippen and Miss Carol Ruth McElroy in carrying out the experiments. The author is grateful to Dr. S. E. Luria for procuring from the Department of Surgery, College of Physicians and Surgeons, Columbia University, the bacteriophage active on the strain of Staphylococcus used in these experiments, as well as for helpful suggestions for improvements in bacteriological technique.

Summary.—In experiments with *Staphylococcus aureus*, strains resistant to penicillin were developed, which retained the property of resistance during the period covered by the experiments. Evidence is presented indicating that resistance is not induced by the action of penicillin on bacteria, but originates through mutation, and that penicillin acts as a selective agent to eliminate nonresistant individuals. Degree of resistance can be increased by exposure to higher concentrations of penicillin, and this increase is interpreted as due to summation of the effects of several independent genetic factors for resistance which undergo consecutive mutation.

* A portion of a research program which is supported in part by a grant from the Josiah Macy, Jr., Foundation.

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THE LAW OF MASS ACTION IN EPIDEMIOLOGY

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Communicated December 13, 1944

Almost all workers in the analytical theory of epidemics assume that the rate at which an infection passes in a population is proportional jointly to the product of the number of persons I who are infectious and the number of persons S who are susceptible to the infection.[1–6] This is called the law of mass action. Thus if the rate of new infections be C the law is written as

$$C = r I S, \qquad (1)$$

where r is a constant. The *a priori* rationalization of the law generally is based upon the assumption, explicit or implicit, that the infectious I are mixing uniformly with the susceptibles S throughout the population. According to the law, if we had a population with twice as many susceptibles and infectious and with the same rate r of mixing, the rate C at which the infection passed would be not twice but four times as great. As a matter of fact, it is unlikely that any such condition exists in detail. For example, it is known that for the childhood infectious diseases such as measles the liability to infection within the family is greater than within the schoolroom and this is in turn greater than that within the community at large. The mixing of the susceptibles and infectious is not uniform throughout the population. Thus the real utility of the assumption for the explanation of the course of an epidemic must be found from the *a posteriori* observation that with the proper choice of a constant r the equation (1) yields a theoretical curve of new cases which is in satisfactory agreement with the observed curve of new cases. Such a value of r is presumably some complicated sort of average value of the different values of r under different degrees of intimacy of contact between different groups of infectious and susceptibles within the population

In the application of (1) the analytical developments vary according to the special assumptions made with respect to the particular disease under consideration. For example, if one is considering malaria and assumes that those once infectious remain so indefinitely and if one neglects accessions to or losses from the population and further neglects the incubation period whether in man or in mosquito, one writes

$$C = \frac{dI}{dt} = r\,I\,S, \qquad S = S_B - I, \tag{2}$$

where S_B is the number of susceptibles at the beginning; then (1) leads to

$$\frac{dI}{dt} = r\,I(S_B - I), \tag{3}$$

which on integration gives

$$I = {}^1/_2 S_B[1 + \tanh {}^1/_2 r S_B(t - t_0)], \tag{4}$$

$$C = {}^1/_4 r S_B{}^2 \operatorname{sech}^2 {}^1/_2 r S_B(t - t_0). \tag{5}$$

This means that the curve of total cases I is the logistic or growth curve, and the curve of new cases is symmetrical with respect to $t = t_0$. The rate of new cases when $t = t_0$ is $rS_B{}^2/4$ and the number of susceptibles remaining at that time is $S_B/2$, half of those at the beginning. In due time all the susceptibles are exhausted.

On the other hand, if the disease is one like measles in which it is generally assumed that there is an incubation period τ and a short period of infectiousness one may write

$$C = rSC(t - \tau) \text{ or } C = (S/m)C(t - \tau), \tag{6}$$

where $m = 1/r$ is the number of susceptibles just sufficient for one infectious case at $t - \tau$ to generate a new infectious case at t. Then, following Soper, and using $C = -dS/dt$ with $u = \log C$, one may obtain, to the order of approximation he uses,

$$\frac{d^2u}{dt^2} = -\frac{r}{\tau} e^u. \tag{7}$$

The integral is

$$u = 2 \log \operatorname{sech} \sqrt{\frac{rC_0}{2\tau}} (t - t_0) + \log C_0 \tag{8}$$

where C_0 is the rate of new cases when $t = t_0$, and then

$$C = C_0 \operatorname{sech}^2 \sqrt{\frac{rC_0}{2\tau}} (t - t_0). \tag{9}$$

It should be noted that the curve of new cases (or, more precisely, the curve of the rate of new cases) is under these assumptions and approximations of the same type as (5) which arose under very different assumptions.

In the third place if one modifies the law of mass action by assuming that the rate of new cases is proportional jointly to some power p of the number of susceptibles[7] and to the case rate lagged by τ, i.e.,

$$C = (S/m)^p C(t - \tau) \tag{10}$$

and eliminates S by $C = -dS/dt$ with $u = \log C$ one finds

$$\frac{d^2u}{dt^2} = -\frac{p}{m\tau} e^{u + (1/p - 1/2)u'} \tag{11}$$

where the first correction term $(1/p - 1/2)(du/dt)$ has been kept in the exponent on the right. This term is neglected by Soper and was neglected, in the analysis above, for $p = 1$; it would appear to be equally negligible for other small values of p, and will be neglected. The integral is then

$$C = C_0 \operatorname{sech}^2 \sqrt{\frac{pC_0}{2m\tau}} (t - t_0), \tag{12}$$

and is still of the same type as (9) and (5). It is therefore clear that the form of the curve of new cases, apart from an interpretation of the constants which are involved and the assumptions which have been made in its derivation, cannot discriminate between a number of different laws of epidemic spread.[8] One could apparently get Soper's equation (7) and an epidemic curve of new cases as the derivative of the growth curve with better approximation from (11) if $p = 2$ than if $p = 1$.

As a matter of fact, one may show directly that when $p = 2$ equation (10) with $C = -dS/dt$ is exactly satisfied by a solution of type (12). For, given

$$\frac{dS}{dt} = \left(\frac{S}{m}\right)^2 \frac{dS}{dt}\bigg|_{t-\tau} \tag{10'}$$

we may substitute therein

$$S = m\left[\cosh \alpha - \sinh \alpha \tanh \alpha \frac{t - t_0}{\tau}\right] \tag{13}$$

and find that the equation is satisfied identically. Then

$$C - \frac{dS}{dt} = \frac{m\alpha}{\tau} \sinh \alpha \operatorname{sech}^2 \alpha \frac{t - t_0}{\tau} \tag{14}$$

and the value of α is connected with the case rate when maximum by

$$C_0 = \frac{m}{\tau} \alpha \sinh \alpha = \frac{m}{\tau}\left[\alpha^2 + \frac{\alpha^4}{6} + \frac{\alpha^6}{120} + \ldots\right] \tag{15}$$

or

$$\alpha = \sqrt{\frac{C_0\tau}{m}}\left[1 - \frac{C_0\tau}{12m} + \frac{29}{1440}\left(\frac{C_0\tau}{m}\right)^2 - \ldots.\right] \tag{15'}$$

For this case the initial and final values of S are

$$S_B = m(\cosh \alpha + \sinh \alpha), \qquad S_E = m(\cosh \alpha - \sinh \alpha)$$

and the value of S at the peak of the epidemic is $S_0 = m \cosh \alpha$ which is halfway between the initial and final values; moreover, $S_B S_E = m^2$ so that the "equilibrium value" m is the geometric mean of the initial and final values of S.

If we return to the general case where $p \neq 2$ and approximations are made in deriving (12) we obtain on integrating (12)

$$S = \text{const} - \sqrt{\frac{2m\tau C_0}{p}} \tanh \sqrt{\frac{pC_0}{2m\tau}} (t - t_0). \qquad (13')$$

The total number of cases from beginning to end of the epidemic is

$$\text{Total cases} = S_B - S_E = 2\sqrt{\frac{2m\tau C_0}{p}} \qquad (16)$$

and to the order of approximation used we find

$$\frac{m}{p} = \frac{(\text{total cases})^2}{8(\text{peak cases})}, \qquad (17)$$

provided we agree to call $C_0\tau$, which is the case rate at the peak of the epidemic multiplied by the incubation interval τ, the "peak cases." Thus what could be determined from an observed epidemic would not be either m or p severally but their ratio. In any such determination it would, of course, be necessary to use the estimated real numbers of total cases and of peak cases and not the total cases or peak cases reported unless the reporting were complete. It should further be observed that actually an epidemic may last over a considerable time and that recruits are coming into the population of susceptibles, which might well make necessary some modification in (17)

Instead of pursuing these considerations at this time we shall turn to the matter of the exact stepwise integration of (10). For notational simplification we introduce as in earlier papers,[6] $x = S/m$, and $T = t/\tau$ so that (10) becomes

$$\frac{dx}{dT} = x^p \left.\frac{dx}{dT}\right|_{-1} \text{ or } \frac{1}{x^p}\frac{dx}{dT} = \left.\frac{dx}{dT}\right|_{-1}. \qquad (10'')$$

The equation may be integrated exactly as[9]

$$\frac{1}{qx_T^q} + x_{T-1} = k = \frac{1}{qx_0^q} + x_{-1}, \qquad (18)$$

where $q = p - 1$ and x_0, x_{-1} are any two values of x which are one incubation period apart. At the beginning and end of the epidemic there are no cases and $x_T = x_{T-1}$. Hence the equation

$$\frac{1}{qx^q} + x - k = 0 \text{ or } \frac{1}{q}e^{-qv} + e^v - k = 0 \qquad (19)$$

with $v = \log x$ will have as solutions the initial and final values x_B, x_E of x or their logarithms. We have

$$f(v) = \frac{1}{q}e^{-qv} + e^{v} - k, \qquad f'(v) = -e^{-qv} + e^{v},$$

$$f'(v) + f'(-v) = 2(\cosh v - \cosh qv).$$

Hence the plot of $f(v)$ has a minimum at $v = 0$, and the (positive) slope for a positive value of v is numerically greater than the (negative) slope for the same numerical but negative value of v, provided $q < 1$, but for $q > 1$ it is less. This means that $v_B + v_K < 0$ or $x_B x_K < 1$ when $q < 1$, i.e., when $0 < p = 2$, but that $x_B x_K > 1$ when $q > 1$, i.e., when $p > 2$.

At the end of the epidemic where the case rates are very small and the values of x are not changing appreciably, C/C_{-1} being x^p is essentially constant and hence the curve of case rates in portions remote from the mode is essentially an exponential curve with a constant difference $p \log x_B$ for the ascending tail and with a constant (negative) difference $p \log x_K$ for the descending tail. The rise will be faster than the fall if $x_B x_K > 1$, i.e., if $p > 2$, but will be slower than the fall if $x_B x_K < 1$, i.e., if $p < 2$. We have seen that for $p = 2$ the curve of case rates is strictly symmetrical. It appears, however, that for $p > 2$ the longer tail would be on the right whereas for $p < 2$ it would be on the left.[10]

[1] Ross, Sir Ronald, "Application of the Theory of Probabilities to the Study of *a priori* Pathometry," Part 1, *Proc. Roy. Soc. London*, **A92**, 204–230 (1915); Ross, Sir Ronald, and Hudson, Hilda P., *Idem.*, Parts 2–3, *Ibid.*, **A93**, 212–240 (1917). The treatment is very general and not limited to the study of epidemics; the law of mass action is introduced under the term "proportional happening"; Part 1, pp. 220 ff.

[2] Lotka, A. J., "Contribution to the Analysis of Malaria Epidemiology," Supplement to *Amer. Jour. Hygiene*, **3**, 1–121 (1923).

[3] Soper, H. E., "The Interpretation of Periodicity in Disease Prevalence," *Jour. Roy. Statist. Soc. London*, **92**, 34–73 (1929).

[4] Frost, W. H., Cutter Lectures, Harvard Medical School, Feb. 2–3, 1928 (unpublished). The method, somewhat adapted, was used in Zinsser, H. and Wilson, E. B., "Bacterial Dissociation and a Theory of the Rise and Decline of Epidemic Waves," *Jour. Prev. Med.*, **6**, 497–514 (1932).

[5] Kermack, W. O., and McKendrick, A. G., "Contributions to the Mathematical Theory of Epidemics," *Proc. Roy. Soc., London*, **115**, 700–721 (1927); **138**, 55–83 (1932); and McKendrick, A. G., "The Dynamics of Crowd Infections," *Edinburgh Med. Jour.*, **47**, 117–136 (1940), where other references are given.

[6] Wilson, E. B., and Burke, M. H., these PROCEEDINGS, **28**, 361–367 (1942); **29**, 43–48 (1943); and Wilson, E. B., and Worcester, J., *Ibid.*, **30**, 37–44 and 264–269 (1944).

[7] A word should be said about the assumption that we might use some power p of S in (10). The assumption may be difficult to justify on *a priori* grounds, but the justification for the case $p = 1$ is none too satisfactory. It would, in fact, be remarkable in a situation so complex as that of the passage of an epidemic over a community if any simple law adequately represented the phenomenon in detail—even to assume that the new case rate should be set equal to any function $f(S)$ of the susceptibles multiplied by the case rate one incubation period earlier might be questioned. We propose to discuss the assumption (10) merely as a possible empirical variant of the case $p = 1$ to see what its

consequences may be. Although mathematics is used to develop the logical inferences from known laws, it may also be used to investigate the consequences of various assumptions when the laws are not known, i.e., one of the functions of mathematical and philosophical reasoning is to keep us alive to what may be only possibilities when the actualities are not yet known.

[8] Ross (Part I, p. 226) develops under the case of "proportional happening" considerations of circumstances which he says "are probably just the conditions which hold in many of the short and sharp epidemics of zymotic diseases, such as measles, scarlatina and dengue." It is perfectly true that the curve of new cases has the form found in epidemics of those diseases, but we have seen that this sort of curve may arise under a variety of different hypotheses. It seems tolerably clear that Ross's theory of happenings, despite its generality, does not include the hypotheses appropriate to the discussion of epidemics of such diseases as measles, for he assumes that his population P has only two divisions, namely, the affected population Z and the susceptible population A and that immunity and the affected condition disappear together. In the case of measles and similar diseases there is a third population, namely, the immunes, let us say Y, so that $P = Z + A + Y$ and his fundamental equations would be replaced by something like:

$$dP = (n - m + i - e)Adt + (N - M + I - E)Zdt + (N' - M' + I' - E')Ydt,$$
$$dA = (n - m + i - e - h)Adt + (N + r)Zdt + (N' + r')Ydt,$$
$$dZ = hAdt + (-M + I - E - r - s)Zdt,$$
$$dY = (-M' + I' - E' - r')Ydt + sZdt.$$

Here n, m, i, e and their correlatives in capitals are natality, mortality, immigration and emigration rates, r is the rate at which the affected return to the susceptibles directly according to Ross's assumption of simultaneous cure and loss of immunity, r' is the rate at which the immunes lose their immunity, s is the rate at which the affected become cured and immune, and h is a factor which under the assumption of proportional happening has the form cZ. These equations do not allow for lag; they assume that births to the affected and to the immunes are susceptible rather than either affected or immune, though for measles children born to the immune mothers are generally themselves immune for a time.

We have tried to reconcile Ross's formulation (which is abstracted by C. O. Stallybrass in his *Principles of Epidemiology*, 1931, pp. 515 ff) and in particular the statement that in infectious diseases the reversion element $rZdt$ implies loss of both immunity and infectiousness, not recovery from disease, by considering the population of affected persons Z to remain affected whether infectious or not as long as they remain immune, but this construction appears impossible. We therefore seem to be forced to the conclusions that Ross's *a priori* pathometry does not cover those zymotic diseases in which immunity with non-infectiousness is a prime phenomenon; it might well cover those in which immunes were permanent carriers, especially if the rates of transfer of infection from the ill and from carriers to susceptibles were not materially different.

[9] If in equation (18) we take $x = 1$ when $T = 0$ and assume $x = 1 + aT + bT^2 + cT^3 + dT^4 + eT^5$, we find, on equating coefficients of powers of T, the following values of b, c, d, e, k in terms of a, good to the power a^5, inclusive.

$$b = pa^2\left[\frac{1}{4} + \frac{p-2}{48}a + \frac{p^2-2p}{192}a^2 + \frac{p^3-2p^2}{768}a^3\right]$$

$$c = pa^2\left[\frac{1}{6} + \frac{7p-6}{72}a + \frac{11p^2-21p+12}{432}a^2 + \frac{97p^3-222p^2+168p}{10368}a^3\right]$$

$$d = pa^3\left[\frac{5p-2}{48} + \frac{8p^2-15p+12}{288}a + \frac{79p^3-186p^2+168p}{6912}a^2\right]$$

$$e = pa^3\left[\frac{p}{30} + \frac{13p^2-24p+24}{1440}a + \frac{35p^3-84p^2+84p}{8640}a^2\right]$$

$$k = \frac{p}{p-1} - a + \frac{p}{12}a^2 - \frac{p^2}{180}a^3 - \frac{13p^3-24p^2+24p}{8640}a^4 - \frac{35p^4-84p^3+84p^2}{51840}a^5.$$

For any assumed slope of the x-curve at $x = 1$ one could plot a short range of that curve, say from $T = -1/2$ to $T = +1/2$. With the value of k one could then proceed stepwise from (18) to any other value of x removed an integral number of units of time for any value assumed within that range.

Particularly interesting, however, is to derive expressions in terms of the maximum case rate $z_0 = C_0\tau/m$. This requires the value of T when $d^2x/dT^2 = 0$, which is

$$T_0 = -\frac{1}{2} + \frac{p-2}{48}a + \frac{5p^2-10p}{576} + \frac{11p^3-26p^2+12p-8}{3072}a^3,$$

and is valid only to the term in a^3 whereas x was valid to the term in a^5. For this value of T, $z = -dx/dT$ takes the value z_0, viz.,

$$z_0 = -a + \frac{p}{8}a^2 - \frac{p^2}{96}a^3 \quad \text{or} \quad a = -z_0 + \frac{p}{8}z_0^2 - \frac{p^2}{48}z_0^3.$$

Hence a may be found in terms of z_0, good to the term in z_0^3, inclusive. With this value of a one may derive expressions in z_0 for k and for the value of x_0 of x at the peak of the epidemic, as follows:

$$k = \frac{p}{p-1} + z_0 - \frac{p}{24}z_0^2 + \frac{p^2}{180}z_0^3, \qquad \text{(A)}$$

$$x_0 = 1 + \frac{1}{2}z_0 - \frac{1}{24}z_0^2 - \frac{113p^2-290p}{11520}z_0^3. \qquad \text{(B)}$$

With these values one may compute stepwise from (18) the values of x for successive values of x removed from the mode by integral numbers of units of time under any assumed value of the ratio $C_0\tau/m$ and for any value of p.

For the approximation that leads to the symmetrical curve, we may write (13′) nearly enough as

$$S = m + \frac{1}{2}C_0\tau - \sqrt{\frac{2\tau m C_0}{p}}\tanh\sqrt{\frac{pC_0}{2\tau m}}(t - t_0). \qquad \text{(C)}$$

Therefore for the beginning and end of the epidemic we should have

$$S_B = m + \frac{1}{2} C_0\tau + \sqrt{\frac{2\tau m C_0}{p}}, \qquad S_E = m + \frac{1}{2} C_0\tau - \sqrt{\frac{2\tau m C_0}{p}}.$$

In general for x at the beginning and end, equation (19) for x may be solved in series. If we set

$$Y = \frac{2}{p}\left(k - \frac{p}{p-1}\right) = \frac{2}{p} z_0 - \frac{1}{12} z_0^2 + \frac{p}{90} z_0^3,$$

$$x = 1 \pm Y^{1/2} + \frac{p+1}{6} Y \pm \frac{(p+1)(2p-1)}{72} Y^{3/2} + \frac{(p+1)(2p-1)(p-2)}{540} Y^2 \pm \frac{(p+1)(2p-1)(2p^2-23p+23)}{17280} Y^{5/2}$$

and x_B will be the value with the positive sign, x_E that with the negative, and

$$\text{Total cases} = 2Y^{1/2} + \frac{(p+1)(2p-1)}{36} Y^{3/2} + \frac{(p+1)(2p-1)(2p^2-23p+23)}{8640} Y^{5/2}$$

provided we measure cases relative to m. Transformed to actual numbers

$$\text{Total cases} = m\left[2Y^{1/2} + \frac{(p+1)(2p-1)}{36} Y^{3/2} + \frac{(p+1)(2p-1)(2p^2-23p+23)}{8640} Y^{5/2}\right], \quad \text{(D)}$$

$$Y = \frac{2}{p}\frac{C_0\tau}{m} - \frac{1}{12}\left(\frac{C_0\tau}{m}\right)^2 + \frac{p}{90}\left(\frac{C_0\tau}{m}\right)^3. \quad \text{(E)}$$

Equation (D) with Y defined as in (E) gives a relationship between total cases, peak cases $C_0\tau$ and m and p which may be solved for any one of those four quantities in terms of the other three to give a more exact expression than (17) which was based on an approximation. If we solve for m and retain only the first approximation beyond (17) we find

$$m = p\frac{(\text{total cases})^2}{8(\text{peak cases})}\left[1 - \frac{5p^2+4p-4}{9p^2}\left(\frac{\text{peak cases}}{\text{total cases}}\right)^2\right]. \quad \text{(F)}$$

For $p \geqq 1$ the correction term in (F) is at most two-thirds of the square of the ratio of peak cases to total cases. This ratio in sharp epidemics of measles (after allowance for under-reporting) is rarely as much as $^1/_6$ so that the correction rarely amounts to more than two percent, and, therefore, considering the difficulty of accurate estimation of actual total cases or peak cases, we may consider (17) a sufficiently good approximation.

[10] The actual calculation of the course of $S = mx$ from (18) for three comparative special cases, with $z = -dx/dT = C/m$, is as follows:

$p = 1, m = 1, z_0 = 0.3$		$p = 2, m = 2, z_0 = 15$		$p = 3, m = 3, z_0 = 10$	
S	Cases	S	Cases	S	Cases
1.9789					
	0.0001				
1.9788		2.9326			
	0.0002		0.0002		
1.9786		2.9324			
	0.0004		0.0002		
1.9782		2.9322		3.9165	
	0.0008		0.0004		0.0003
1.9774		2.9318		3.9162	
	0.0016		0.0008		0.0006
1.9758		2.9310		3.9156	
	0.0032		0.0018		0.0015
1.9726		2.9292		3.9141	
	0.0063		0.0038		0.0033
1.9663		2.9254		3.9108	
	0.0123		0.0084		0.0072
1.9540		2.9170		3.9036	
	0.0238		0.0178		0.0156
1.9302		2.8992		3.8880	
	0.0455		0.0368		0.0336
1.8847		2.8624		3.8544	
	0.0837		0.0732		0.0694
1.8010		2.7892		3.7848	
	0.1446		0.1356		0.1320
1.6564		2.6536		3.6528	
	0.2231		0.2190		0.2175
1.4333		2.4346		3.4353	
	0.2866		0.2864		0.2865
1.1467		2.1482		3.1488	
	0.2857		0.2864		0.2865
0.8610		1.8618		2.8623	
	0.2140		0.2190		0.2205
0.6470		1.6428		2.6418	
	0.1246		0.1356		0.1389
0.5224		1.5072		2.5029	
	0.0612		0.0732		0.0768
0.4612		1.4340		2.4261	
	0.0274		0.0366		0.0396
0.4338		1.3974		2.3865	
	0.0117		0.0178		0.0198
0.4221		1.3796		2.3667	
	0.0049		0.0084		0.0096
0.4172		1.3712		2.3571	
	0.0021		0.0038		0.0048
0.4151		1.3674		2.3523	
	0.0009		0.0018		0.0024
0.4142		1.3656		2.3499	
	0.0003		0.0008		0.0009
0.4139		1.3648		2.3490	
	0.0002		0.0004		0.0006
0.4137		1.3644		2.3484	
			0.0002		0.0003
		1.3642		2.3481	
			0.0002		
		1.3640			

The left-hand skewness for $p = 1$ and right-hand skewness for $p = 3$ show in the figures. The different values of z_0 were chosen so that the case rates at maximum $C_{0T} = z_0 m$ and total cases should be the same provided the approximate formula (16) were used and the value of x_0 were taken from (B) in footnote 9. Slight irregularities in the numbers must be expected due to the limited number of places carried, and slight discrepancies in verifying (16) from the calculations because of the approximative nature of (16) and (B).

J. Brownlee stated, *Proc. Roy. Soc. Med., Epid. Sect.*, **2**, Part 2, 243–258 (1909): . . . the symmetry of the course of the epidemic is an obvious and marked feature. The deduction from this phenomenon is direct and complete, namely, that the want of persons liable to infection is not the cause of the decay of the epidemic. On no law of infection which I have been able to devise would such a cause permit epidemic symmetry. The fall must in all cases be much more rapid than the rise, though, on the contrary, when asymmetry is markedly present the opposite holds. Ross[1] comments on this statement. We may point out that if we accept the generalization of the law of mass action suggested in (10) there is symmetry for $p = 2$, negative skewness for $p < 2$ and positive skewness for $p > 2$. Thus a rather simple law has been devised which may explain symmetry or skewness of either sign. Furthermore, in the examples above which correspond to rather severe epidemics of measles the rise is at the (logarithmic) rate $p \log x_B$ or 0.68, 0.77, 0.81, respectively, for $p = 1, 2, 3$; and the rate of the fall is $-p \log x_E$ or 0.89, 0.77, 0.74, respectively. In the first case the rate of fall is considerably greater than the rate of rise, in the second case they are equal, and in the third case the rate of fall is but slightly less than the rate of rise. With higher values of p the rate of fall would become considerably less than the rate of rise, but even with very high values of p and with the same values of peak cases and of total cases as in the illustrations above the rate of rise could probably not exceed 0.89 and the rate of fall not be lower than 0.69.

A LETTER FROM LORD RAYLEIGH TO J. WILLARD GIBBS AND HIS REPLY

BY EDWIN B. WILSON

HARVARD SCHOOL OF PUBLIC HEALTH

Communicated December 4, 1944

In the small collection of letters left by J. W. Gibbs and now in the possession of Ralph G. Van Name is one from Lord Rayleigh the answer to which I presumed still existed because the present Lord Rayleigh quoted three sentences from it in his biography of his father.[1] When I sent a copy of his father's letter to Lord Rayleigh, he kindly sent me a transcript of Gibbs's reply. As this exchange of letters between a foreign associate and a member of this Academy seems to me likely to be of sufficient interest to our members and of sufficient importance to the history of science to justify publication in full even at this late date, I have secured the permission of Lord Rayleigh and of Professor Van Name to print them here.

June 5/92
Terling Place,
Witham, Essex.

Dear Prof. Gibbs:

I have been experimenting lately upon the intensity of reflection from water at nearly perpendicular incidence, and the not very close agreement that I have found with Young's formula $\left(\frac{\mu - 1}{\mu + 1}\right)^2$ has set me thinking whether there is any reason for expecting this formula to be correct, when the effect of *dispersion is included*. The case of two strings joined together one (as usual) perfectly flexible and the other with such stiffness as to introduce dispersion, shews that the reflection is *not* to be got from $\frac{\mu - 1}{\mu + 1}$, even though the correct μ be used; I think the deviation from this is of the same order as the deviation of $\frac{\mu_\lambda - 1}{\mu_\lambda + 1}$ from $\frac{\mu_\infty - 1}{\mu_\infty + 1}$. But this case does not much resemble optical dispersion (one would suppose), and I am writing partly to ask whether you have ever considered the problem of reflection with inclusion of dispersion on the lines of your published papers. And with respect to the latter I find a difficulty in your estimation on Kelvin's theory [is not this analytically identical with Lorenz?] of the potential energy as

$$\frac{\pi^2 B h^2}{l^2} + \frac{b h^2}{4}$$

(*Phil. Mag.*, XXVII, p. 24) in which the 1st contains l and the 2nd does not. Supposing the disturbance from uniformity to be by simple changes of elastic quality, would not the 2nd term contain l^2 like the first.

And now on another subject. Have you ever thought of bringing out a new edition of, or a treatise founded upon, your "Equilibrium of Het. Substances." The original version though now attracting the attention it deserves, is too condensed and too difficult for most, I might say all, readers. The result is that as has happened to myself, the idea is not grasped until the subject has come up in one's own mind more or less independently. I am sure that there is no one who could write a book on Thermodynamics like yourself.

I feel that I am taking a liberty in writing like this, but it is in the interest of science and you will forgive me.

I remain
yrs very truly
Rayleigh

New Haven.
June 27, 1892.

My dear Lord Rayleigh,

The electrical theory of light seems to afford a very simple equation for harmonic motions of any one period in any optical field, *without* neglect of the causes of dispersion. If we consider an element of space containing

many ponderable molecules, the electrical motions in that space are presumably very complicated, but they result from the motions at a distance, and the effect of motions at a distance can be expressed by a very simple formula. There is first the electromotive force of induction, of which the components are calculated from the components of acceleration by the same law as the potential is calculated in the theory of gravitation from the density of matter. If we write Pot to express this operation, we will have $-\text{Pot}\,\ddot{F}$ for this force, F being a bi-vector representing the displacement (*i. e.*, the three components of F are complex realms). There is also (or may be) an electrostatic force which can be represented by ∇Q, where Q is a complex realm (the electrostatic potential). It should be borne in mind that the displacements which occur in the equations of wave motion are a sort of average for elements of space which are large in comparison with the distances of neighboring molecules, but small in comparison with a wave-length. If F is understood as representing such an average, and likewise Q

$$-\text{Pot}\,\ddot{F} - \nabla Q$$

will still correctly represent the electromotive forces acting upon the element from a distance. Whether this formula correctly represents the internal forces of the element or those forces which arise from the immediate neighbourhood of the element is of no consequence. Now I say that F is a function of $-\text{Pot}\,\ddot{F} - \nabla Q$. This is really saying very little. It is only saying that if under any circumstance an average harmonic motion F subsists in an element of space, and if we then change things in the remoter parts of the field, but so that the value of $-\text{Pot}\,\ddot{F} - \nabla Q$ shall remain unaltered at the element considered, the same motion F will continue to subsist in that element. F is therefore a function of $-\text{Pot}\,\ddot{F} - \nabla Q$ and by the principle of superposition of motions a linear function. Or, we may say that $-\text{Pot}\,\ddot{F} - \nabla Q$ is a linear function of F, and write as in my paper on Kelvin's quasi-labile ether.

$$-\text{Pot}\,\ddot{F} - \nabla Q = 4\pi\Phi F$$

the operator Φ representing in general a linear vector function, which however in the case of an isotropic body reduces to a (so-called) numerical quantity (*real*, if the body is transparent). It will of course vary in different parts of the field with the optical properties of the bodies in the field. The equation is not indeed absolutely accurate—the definitions on which it is founded are not absolutely sharp, and the phenomena of (non-magnetic) rotation of plane of polarization form a striking exception to the equation, which I have discussed at length in my 2nd "Note on the Elec. Theory of Light." We may say that the inaccuracy in the equation results from the virtual assumption that the structure of a body is infinitely fine as measured by a wave-length of light.

But I do not see that any amount of *dispersion* constitutes any reason for the failure of the equation. It will apply, so far as I can see, even to cases of selective absorptive and abnormal (so-called) dispersion, as well as to any degree of opacity.

Now this general equation (as I have pointed out in my paper on Kelvin's

quasi-labile ether) gives us on one hand Fresnel's wave-surface, and on the other Fresnel's laws of intensities of reflected light, if we suppose that Φ is constant on each side of a mathematical plane of reflection, which is of course a very precarious supposition. You will remember that in one of your papers you refer to one of Lorentz, in which he claims to obtain a remarkable agreement with Jamin's experiments for solids by supposing the change in index (i.e., in Φ) to be gradual. Your own experiments have shown that at least in the case of liquids the influence of foreign substances at the surface is considerable. Besides the question of the gradual or abrupt change in Φ there is the question (at least when the foreign substances are present) of values of Φ which are not intermediate, as complex values where Φ is real on each side of the surface.

I was very glad that you vindicated Fresnel's law from the most considerable deviation as reported by Jamin for liquids, and feel quite sorry that your later experiments show in another way a deviation. I should hope for the sake of the theory (to which you see I am somewhat attached) that the deviations can be accounted for by attributing some not unnatural qualities to a thin film at the plane of reflection.[2]

With respect to reflection of waves in strings, we may cause dispersion by supporting a heavy string by threads (very close together) from the ceiling. For waves of any one period (in time) the reflection where two such strings are joined is determined entirely by the wave-lengths in the two strings.

I am afraid that I hardly have the right to say what the potential energy should be on Kelvin's theory, in respect to a point on which he has not expressed himself. It is however natural, if we imagine a sea of jelly to be surging to and fro among fixed molecules, to suppose that the jelly is thereby distorted. There are indeed two exceptions; 1st. If we suppose that the molecules do not affect the motion of the jelly, although they may have made it denser (by their attraction); 2nd. If we suppose the molecules to move with the jelly, taking the same displacements. In either of these cases (both apparently improbable) the momentum of the jelly would have the same ratio to the velocity for all directions about a point. This is not the case in Kelvin's theory, in which the effective inertia is aeolotropic (*Phil. Mag.* XXVI, 501). If then the jelly is distorted in its motion by the molecules among which it flows, the potential energy should not vanish for $l = \infty$, at least apparently not. (It is hard to speak with absolute certainty in regard to so peculiar a jelly, which enjoys moreover, I suppose, the privilege of further hypotheses in its behalf as they may become necessary.) I think we may say that it is reasonable to admit such a term in the formula on *a priori* evidence, its magnitude (whether zero or otherwise) to be determined *a posteriori*. I am not claiming the existence of an absolutely constant term, but one which does not vanish for $l = \infty$. I should expect, however, that it would be tolerably constant in most cases. As a matter of fact, it seems to me that we can measure it, as compared with the other term, by means of the dispersion of light, as I have in-

dicated in the passage in question. As thus measured, it does not appear at all constant (for constant amplitude) as I should expect it to be. This seems to me a very serious objection to the theory, although one which I should urge with great diffidence, as Lord Kelvin has not touched upon these points.

I thank you very much for your kind interest in my "Equilib. Het. Subst." I myself had come to the conclusion that the fault was that it was too *long*. I do not think that I had any sense of the value of time, of my own or others, when I wrote it.[3] Just now I am trying to get ready for publication something on thermodynamics from the *a priori* point of view, or rather on "Statistical Mechanics" of which the principle interest would be in its application to thermodynamics—in the line therefore of the work of Maxwell and Boltzmann. I do not know that I shall have anything particularly new in substance, but shall be contented if I can so choose my standpoint (as seems to me possible) as to get a simpler view of the subject.

I remain
Your's faithfully
J. Willard Gibbs

[1] *Life of Lord Rayleigh*, London, 1924. See pp. 172–173.

[2] As I interpret the statement of Lord Rayleigh in his *Scientific Papers*, vol. IV, p. 12, where the results of his experiment in question are reprinted, he finally concluded that the discrepancy between theory and experiment was not sufficient to require any explanation.

[3] This and the preceding two sentences are the three quoted in the Life of Lord Rayleigh. The reference to Statistical Mechanics which immediately follows is of particular interest in view of the publication about a decade later of the Elementary Principles in Statistical Mechanics.

STUDIES ON GEODESICS IN VIBRATIONS OF ELASTIC BEAMS

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Communicated December 12, 1944

The transverse vibrations of a homogeneous elastic beam that is simply supported (hinged) at both ends satisfy the fourth order partial differential equation

$$\frac{\partial^2 u(x, t)}{\partial t^2} + c^2 \frac{\partial^4 u(x, t)}{\partial x^4} = 0 \qquad (1)$$

and the boundary conditions

$$u(o, t) = \frac{\partial^2 u(x, t)}{\partial x^2}\Big]_{x=0} = 0, \; u(l, t) = \frac{\partial^2 u(x, t)}{\partial x^2}\Big]_{x=l} = 0, \qquad (2)$$

where the constant $c^2 = EI/\rho$, the ratio of the flexural rigidity EI and mass ρ per unit length of the beam, and l is the length of the beam.[1]

The kinetic and potential energies are given, respectively, by

$$T = \frac{\rho}{2}\int^{l}\left(\frac{\partial u\,(x,\,t)}{\partial t}\right)^2 dx,\; U = \frac{EI}{2}\int_0^{l}\left(\frac{\partial^2 u(x,\,t)}{\partial x^2}\right)^2 dx. \tag{3}$$

It is well known that the differential equation (1) is the Euler-Lagrange equation resulting from *Hamilton's Principle*

$$\delta\int_{t_0}^{t_1}\Big(T - U\Big)dt = 0 \tag{4}$$

when the usual restrictions are made on fourth order derivatives. There is a conservation of total energy so that $T + U = C$, a constant along a motion of the beam.

A one-parameter family of positions of the vibrating beam will be called a *dynamical path* or *dynamical trajectory*. A dynamical path need not be specified by the time parameter t. Let $u = u(x, t)$ be a dynamical path with the time parameter t and total energy C. Let us define a new parameter s along this dynamical path by

$$s = \int_0^{t} A(\lambda)d\lambda, \tag{5}$$

where

$$A(\lambda) = 2C - EI\int_0^{l}\left(\frac{\partial^2 u(x,\,\lambda)}{\partial x^2}\right)^2 dx. \tag{6}$$

In terms of this new parameter s, the conservation of total energy yields the condition

$$\rho\int_0^{l}\left(\frac{\partial\bar{u}(x,\,s)}{\partial s}\right)^2 dx = \frac{1}{\bar{A}(s)} \tag{7}$$

on defining $\bar{u}(x,\,s) = u(x,\,t(s))$ and $\bar{A}(s) = A(t(s))$. Now condition (7) can be written in the following equivalent form:

$$\bar{A}^2(s)\frac{\rho}{2}\int_0^{l}\left(\frac{\partial\bar{u}(x,\,s)}{\partial s}\right)^2 dx + \frac{EI}{2}\int_0^{l}\left(\frac{\partial^2\bar{u}(x,\,s)}{\partial x^2}\right)^2 dx = C. \tag{8}$$

This then is the statement of the conservation of total energy when it is expressed in terms of the parameter s instead of the time t.

If the boundary conditions (2) are used in integrations by parts and if we employ the energy condition (7) or (8), we can establish the following theorem.

THEOREM 1. *If $u = u(x\ t)$ is a dynamical path with the time parameter t and total energy level C, then in terms of the parameter s defined in (5), it satisfies the integro-differential equation*

$$\frac{\partial^2\bar{u}(x,s)}{\partial s^2} + \frac{1}{\bar{A}(s)}\left\{-2EI\left[\int_0^l \frac{\partial^4\bar{u}(x_1,s)}{\partial x_1^4}\,\frac{\partial\bar{u}(x_1,s)}{\partial s}\,dx_1\right]\frac{\partial\bar{u}(x,s)}{\partial s} + EI\left[\int_0^l \left(\frac{\partial\bar{u}(x_1,s)}{\partial s}\right)^2 dx_1\right]\frac{\partial^4\bar{u}(x,s)}{\partial x^4}\right\} = 0. \quad (9)$$

In other words, the partial differential equation (1) for the vibration states of a beam, simply supported at both ends and having *the same total energy level C*, becomes the integro-differential equation (9) when it is expressed in terms of the parameter *s*.

Let us now consider a converse problem to that of deriving (9). In fact, we shall start with the equation (9) and *assume* that $\bar{u}(x, s)$ is a solution of (9) with certain properties. If we define a parameter t by

$$t = \int_0^s \frac{d\lambda}{\bar{A}(\lambda)}, \quad (10)$$

where

$$\bar{A}(\lambda) = 2C - EI\int_0^l \left(\frac{\partial^2\bar{u}(x,\lambda)}{\partial x^2}\right)^2 dx, \quad (11)$$

then one can prove the following result.[2]

THEOREM 2. *If $\bar{u}(x, s)$ is a solution of the integro-differential equation (9) such that $\bar{A}(s) > 0$, $\bar{u}(o, s) = \bar{u}(l, s) = 0$, $\left.\frac{\partial^2\bar{u}(x,s)}{\partial x^2}\right]_{x=0} = \left.\frac{\partial^2\bar{u}(x,s)}{\partial x^2}\right]_{x=l} = 0$, if s is a general parameter, C is a fixed positive number, and if $\bar{u}(x, s)$ satisfies the energy integral (7), or equivalently (8), then by the change (10) of parameter s to the time parameter t, the function $u(x, t)$ defined by $u(x, t) = \bar{u}(x, s(t))$ will satisfy the partial differential equation (1), the boundary conditions (2) and will have a total energy level C—in other words $u = u(x, t)$ will be a dynamical path in the time parameter t and will have a total energy level C.*

To bring out some geometrical ideas connected with our subject, let us multiply both sides of the energy integral (7) by $\bar{A}(s)\left(\frac{ds}{d\lambda}\right)^2$, where λ is an arbitrary (suitably admissible) parameter. This gives

$$2\left[C - \frac{EI}{2}\int_0^l \left(\frac{\partial^2\bar{u}(x,s(\lambda))}{\partial x^2}\right)^2 dx\right]\rho\int_0^l \left(\frac{\partial\bar{u}(x_1,s(\lambda))}{\partial\lambda}\right)^2 dx_1 = \left(\frac{ds}{d\lambda}\right)^2. \quad (12)$$

Hence, if we define $v(x, \lambda)$ by $v(x, \lambda) = \bar{u}(x, s(\lambda))$ we obtain

$$s = \int_{\lambda_0}^{\lambda} \left\{ 2\left[C - \frac{EI}{2}\int_0^l \left(\frac{\partial^2 v(x, \lambda_1)}{\partial x^2}\right)^2 dx \right] \rho \int_0^l \left(\frac{\partial v(x_1, \lambda_1)}{\partial \lambda_1}\right)^2 dx_1 \right\}^{1/2} d\lambda_1. \tag{13}$$

The parameter s defined by (13) is, however, the arc length of a curve $v = v(x, \lambda_1)$, parameterized with parameter λ_1, $(\lambda_0 \leq \lambda_1 \leq \lambda)$, in an *infinitely dimensional "Riemannian" geometry* with function coordinates $v(x)$ and element of arc length squared given by

$$ds^2 = 2\left[C - \frac{EI}{2}\int_0^l \left(\frac{d^2 v(x)}{dx^2}\right)^2 dx \right] \rho \int_0^l \left(\delta v(x_1)\right)^2 dx_1, \tag{14}$$

where $\delta v(x)$ as well as $v(x)$ are arbitrary continuous functions with fourth order continuous derivatives such that $v(x)$ and $\delta v(x)$ as well as their second derivatives vanish at both ends of the beam, $x = 0$ and $x = l$. It is important to notice here that *the boundary conditions for a beam hinged at both ends play an important rôle in the determination of the function coordinate space of the geometry.*[3] We shall assume that $v(x)$ satisfies the inequality

$$C > \frac{EI}{2}\int_0^l \left(\frac{d^2 v(x)}{dx^2}\right)^2 dx,$$

a property that guarantees the positive definiteness of the quadratic functional differential form (14).

By definition, the curves $v = v(x, \lambda)$ that make the length functional (13) have a stationary value are the *geodesics* of our infinitely dimensional "Riemannian" space. The generalized Euler-Lagrange equation for this generalized calculus of variations problem can be shown to be the *integro-differential equation* (9) provided that the general parameter λ_1, is taken to be the arc length parameter s and provided $\bar{u}(x, s)$ is replaced throughout by $v(x, s)$.

If λ is the time t, $v(x, t)$ is a solution of the beam problem hinged at both ends and having total energy C, and if $T(t)$ is the *corresponding* kinetic energy at any time t, then it can be shown readily that the arc length s of the dynamical path $v = v(x, t)$ is given by

$$s = \int_0^t 2T(t_1)dt_1.$$

This result provides a physical interpretation for the arc length of a geodesic in our infinite dimensional "Riemannian" space. From another point of view, it can be regarded as a *generalization of Hamilton's characteristic function* [4] Hamilton introduced his characteristic function (action integral) in his studies on optics and then later in his studies on dynamical systems with a finite degree of freedom and with an energy integral.

It is clear from our previous discussion that the energy integral (8) for a solution of the integro-differential equation (9) is equivalent to the demand that the parameter s be an arc length parameter of the infinite dimensional "Riemannian" space (14). With the aid of this result, we can summarize Theorem 1 and Theorem 2 briefly in the language of vibrations as follows:

THEOREM 3. *The vibration states (dynamical path) with total energy level C of an elastic beam with both ends hinged can be represented by a geodesic in the infinite dimensional "Riemannian" space whose element of arc length squared is given by (14), and conversely.*

It can be shown that the infinite dimensional "Riemannian" space (14) is *not* of constant "Riemannian" curvature.[5] Hence the *vibrations of an elastic beam hinged at both ends with constant total energy furnish a physical model for the geodesics of a special type of infinite dimensional "Riemannian" space with variable "Riemannian" curvature.*

The harmonic vibrations of the simply supported beam are given by

$$u_n(x, t) = D_n \sin \frac{n\pi x}{l} \sin w_n t \ (n = 1, 2, \ldots, \ldots),$$

where the angular frequency

$$w_n = \frac{n^2\pi^2}{l^2}\sqrt{\frac{EI}{\rho}}.$$

A simple calculation shows that for $C > 0$ and for each integral value of n, the harmonic vibration

$$u_n(x, t) = \frac{2}{n^2\pi^2}\sqrt{\frac{l^3C}{EI}} \sin \frac{n\pi x}{l} \sin\left(\frac{n^2\pi^2}{l^2}\sqrt{\frac{EI}{\rho}}t\right)$$

is a closed geodesic in the infinite dimensional "Riemannian" space with element of arc length ds given by (14). It can also be shown that this infinite system of closed geodesics $u_1(x, t)$, $u_2(x, t)$, . . . , . . . forms a mutually orthogonal system of curves at the origin $v(x) = 0$ of the space (14) and that the tangent vectors $\frac{\partial u_n(x, t)}{\partial t}$ to these geodesics at the origin have the same magnitude given by twice the total energy C of each of these harmonic vibrations.

With the same general methods and obvious occasional changes in detail, one can treat the vibrations of cantilever beams, the vibrations of other beams, and many other problems in the mechanics of continuous media.[6]

[1] See Kármán and Biot, *Mathematical Methods in Engineering*, pp. 267–273, 283–290. See also Timoshenko, S., *Vibration Problems in Engineering*, p. 221.

[2] The method of proof and details are similar to those given by the author in another connection. See Michal, A. D., *The Vibrations of Elastic Strings as Studies in Geodesics.*

[3] The boundary conditions of functional equations play a similar geometrical rôle in many other connections. In the applications, this is not only the case in all sorts of vibration problems, but also in many other physical phenomena.

[4] See Whittaker, E. T., *Analytical Dynamics* (Cambridge, 1927), pp. 289 and 317. The reader who is acquainted with the Maupertuis-Jacobi "Least" Action Principle in finite degrees of freedom will notice that we have incidentally given a generalization of this principle for an infinite degree of freedom problem.

[5] The general tensor calculus and the geometric concepts associated with it can be improvised by anyone who is acquainted with the author's general "Riemannian" geometries as studied in Michal, A. D., "General Differential Geometries and Related Topics," *Bull. Amer. Math. Soc.*, **45** 529–563 (1939), especially pp. 551–559. References to the author's earlier work are also given in this paper.

[6] While giving an account of this paper in a lecture to my Seminar on Applied Mathematics, Bateman kindly called my attention to an application of Haar's work on the calculus of variations to Hamilton's Principle (4). Assuming only the existence of continuous second order partial derivatives, Hamilton's Principle leads to the *two* partial differential equations

$$\frac{\partial u(x, t)}{\partial t} = c^2 \frac{\partial^2 w(x, t)}{\partial x^2}, \quad \frac{\partial^2 u(x, t)}{\partial x^2} = -\frac{\partial w(x, t)}{\partial t} \tag{a}$$

in terms of an auxiliary function $w(x, t)$. If continuous fourth order derivatives are assumed to exist, the elimination of $w(x, t)$ in (a) leads to the classical equation (1) for beam vibrations. In terms of the parameter s, equations (a) become the *two integro-differential equations*

$$\frac{\partial \bar{u}(x, s)}{\partial s} = \frac{c^2}{\bar{A}(s)} \frac{\partial^2 \bar{w}(x, s)}{\partial x^2}, \quad \frac{\partial \bar{w}(x, s)}{\partial s} = -\frac{1}{\bar{A}(s)} \frac{\partial^2 \bar{u}(x, s)}{\partial x^2} \tag{b}$$

If continuous fourth order derivatives are assumed to exist, the elimination of $\bar{w}(x, s)$ in (b) leads to the fundamental integro-differential equation (9).

Similar remarks can be made in other problems such as the vibrations of an elastic string.

AN EXTENSION OF LIE'S THEOREM ON ISOTHERMAL FAMILIES

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Communicated December 7, 1944

1. Conformal Representation.—Let a surface Σ be represented conformally upon a plane π with cartesian coordinates (x, y). The linear-element of Σ is then of the form

$$ds^2 = E(x, y)(dx^2 + dy^2), \tag{1}$$

where $E(x, y) > 0$. The parametric curves $x = \text{const.}$ and $y = \text{const.}$ form an isothermal net on Σ.

It is well known that $h(x, y) = c$, where h is a harmonic function of (x, y), that is, h satisfies the Laplace equation, $h_{xx} + h_{yy} = 0$, defines an isothermal family of curves on Σ. The constant c is called the isothermal parameter.

The converse of the preceding statement is not valid. That is, if $g(x, y) = \text{const.}$ defines an isothermal family of curves on Σ with linear-element of the form (1), then it does not follow necessarily that g is a harmonic function, but it must be a function of a harmonic function. The exact statement which is due to Lie is as follows. The family of curves $g(x, y) = \text{const.}$ represents an isothermal system on the surface Σ with linear-element of the form (1), if and only if g satisfies the partial differential equation of third order

$$\left(\frac{\partial^2}{\partial x^2} + \frac{\partial^2}{\partial y^2}\right) \arctan \frac{g_y}{g_x} = 0. \tag{2}$$

This means geometrically that the angle θ between the ∞^1 curves $g(x, y) = \text{const.}$ and the isothermal family $x = \text{const.}$ (or $y = \text{const.}$) is a harmonic function of (x, y).

2. Statement of Our Problem.—We propose to give the necessary and sufficient condition that $g(x, y) = \text{const.}$, represent an isothermal family upon a surface Σ when (x, y) are general curvilinear coordinates on Σ. That is, when the linear-element of Σ is of the form

$$ds^2 = E(x, y)dx^2 + 2F(x, y)dxdy + G(x, y)dy^2, \tag{3}$$

where $H^2 = EG - F^2 > 0$, we shall determine the necessary and sufficient condition that $g(x, y) = \text{const.}$ represents an isothermal system of curves on Σ. This leads to a wide extension of the theorem of Lie, which is stated above.

The preceding condition is simpler when the parametric curves form an orthogonal net, that is, when $F = 0$. Of course, the simplest condition is (2) when the parametric curves form an isothermal net and (x, y) are isothermal coordinates.

As an application of our preceding work, we shall obtain the necessary and sufficient condition that the ∞^1 curves $g(x, y) = \text{const.}$ shall represent an isothermal family upon the Monge surface $\Sigma: z = f(x, y)$.

Finally the condition is found that the level curves $z = \text{const.}$ of the surface $\Sigma: z = f(x, y)$ be an isothermal family. This is applied to the mapping upon a plane π of the loxodromes (relative to the level curves) of the surface Σ, showing that they can be represented by straight lines for a sphere (Mercator) and a spheroid (Lambert), (and for any minimal surface and also for any surface of revolution with axis perpendicular to the xy-plane), but not for an ellipsoid of three unequal axes. Here use is made of a theorem of Kasner, which states that the complete system of ∞^2 isogonal trajectories of a given family of curves is linear if and only if the given family is isothermal.

3. The Condition When the Minimal Lines Are Given in the Finite Form.—Let (u, v) denote the minimal coordinates of any point on the surface Σ. Then $u = x + iy$, $v = x - iy$ where (x, y) are the isothermal coordinates defined in (1).

If a general point transformation is applied to the plane π upon which the surface Σ is conformally represented by means of the equation (1), it is found that u and v are given by the general expressions

$$u = \phi(x, y), \qquad v = \psi(x, y), \tag{4}$$

where ϕ and ψ are conjugate complex functions of the real variables (x,y). Of course, (x, y) are now general curvilinear coordinates of any point on Σ since the linear-element of Σ is of the form (3). The finite forms of the equations of the minimal lines are $\phi(x, y) = \text{const.}$ and $\psi(x, y) = \text{const.}$

We seek the condition for an isothermal family in the general curvilinear coordinates (x, y). According to Lie's theorem, any isothermal family is defined in minimal coordinates (u, v) by a differential equation of the form

$$\log \frac{dv}{du} = \lambda(u) + \mu(v). \tag{5}$$

By substituting (4) into this equation, we find that the ∞^1 curves defined by the differential equation of first order $dy/dx = p = p(x, y)$ form an isothermal family if and only if the function p of (x, y) satisfies an equation of the form

$$\log \frac{\psi_x + p\psi_y}{\phi_x + p\phi_y} = \lambda(\phi) + \mu(\psi). \tag{6}$$

The functions λ of ϕ and μ of ψ must be eliminated by partial differentiation. Firstly upon applying the operation $\phi_y\partial/\partial x - \phi_x\partial/\partial y$ and simplifying, and secondly the operation $\psi_y\partial/\partial x - \psi_x\partial/\partial y$ to the above equation, we obtain the expression

$$\left(\psi_y \frac{\partial}{\partial x} - \psi_x \frac{\partial}{\partial y}\right)\left[\frac{(\phi_y(\partial/\partial x) - \phi_x(\partial/\partial y))\log \dfrac{\psi_x + p\psi_y}{\phi_x + p\phi_y}}{\phi_x\psi_y - \phi_y\psi_x}\right] = 0. \quad (7)$$

This condition is the necessary and sufficient condition that the curves defined by the differential equation $dy/dx = p(x, y)$ form an isothermal family on the surface Σ whose minimal curves are given in the finite form by $\phi(x, y) =$ const. and $\psi(x, y) =$ const.

4. The Condition (7) in Terms of the Differential Equations of the Minimal Curves.—For this purpose, let us define $\alpha(x, y)$ and $\beta(x, y)$ by the equations

$$\phi_x = -\alpha\phi_y, \quad \psi_x = -\beta\psi_y,$$

so that the differential equations of the minimal curves are $dy/dx = \alpha$ and $dy/dx = \beta$.

Upon substituting these into (7) and simplifying, we obtain the expression ultimately

$$\left[\frac{\partial^2}{\partial x^2} + (\alpha + \beta)\frac{\partial^2}{\partial x\partial y} + \alpha\beta\frac{\partial^2}{\partial y^2}\right]\log\frac{p - \beta}{p - \alpha} + \left(\frac{\beta_x + \alpha\beta_y}{\alpha - \beta}\right)\times$$

$$\left(\frac{\partial}{\partial x} + \alpha\frac{\partial}{\partial y}\right)\log\frac{p - \beta}{p - \alpha} - \left(\frac{\alpha_x + \beta\alpha_y}{\alpha - \beta}\right)\left(\frac{\partial}{\partial x} + \beta\frac{\partial}{\partial y}\right)\log\frac{p - \beta}{p - \alpha} +$$

$$(\beta\alpha_{yy} - \alpha\beta_{yy} + \alpha_{xy} - \beta_{xy}) + \left(\frac{\alpha_y - \beta_y}{\alpha - \beta}\right)(\alpha\beta_y - \beta\alpha_y + \beta_x - \alpha_x) = 0. \quad (8)$$

This is the necessary and sufficient condition that the family of curves defined by the differential equation $dy/dx = p = p(x, y)$ is an isothermal family when the minimal curves of the surface Σ are given by the differential equations $dy/dx = \alpha(x, y)$ and $dy/dx = \beta(x, y)$.

5. The Condition (8) in Terms of the General Form (3) of the Linear-Element of the Surface Σ.—If (x, y) are curvilinear coordinates on the surface Σ such that the linear-element of Σ is given by the general equation (3), it follows that α and β must be given by

$$\alpha = \frac{-1}{G}(F - iH), \qquad \beta = -\frac{1}{G}(F + iH). \quad (9)$$

Let θ denote the expression

$$\theta = \text{arc cot } \frac{1}{H}(pG + F). \tag{10}$$

This is actually the angle θ between any curve of the family $dy/dx = p(x, y)$ and the parametric curves $x = \text{const}$.

Substituting the values of α and β as given by equations (9), we find that the equation (8) assumes the form

$$H\left[G\frac{\partial^2\theta}{\partial x^2} - 2F\frac{\partial^2\theta}{\partial x\partial y} + E\frac{\partial^2\theta}{\partial y^2}\right] + [HG_x - GH_x + FH_y - HF_y]\frac{\partial\theta}{\partial x} +$$

$$[FH_x - HF_x + \frac{1}{G}\{-EHG_y + 2FHF_y + (EG - 2F^2)H_y\}]\frac{\partial\theta}{\partial y} +$$

$$\frac{H}{G}[HF_{yy} - FH_{yy} + GH_{xy} - HG_{xy}] + \frac{1}{G^2}(GH_y + HG_y)(FH_y - HF_y) +$$

$$\frac{1}{G^2}(H^2G_xG_y - G^2H_xH_y) = 0. \tag{11}$$

This is our extension of Lie's theorem on isothermal familes. If it is desirable to write the above equation in a form which does not contain partial derivatives of H, we find the form

$$2H^2\left[G\frac{\partial^2\theta}{\partial x^2} - 2F\frac{\partial^2\theta}{\partial x\partial y} + E\frac{\partial^2\theta}{\partial y^2}\right] + [(EG-2F^2)G_x - G^2E_x + 2FGF_x +$$

$$EFG_y + GFE_y - 2EGF_y]\frac{\partial\theta}{\partial x} + [EFG_x + GFE_x - 2EGF_x - E^2G_y +$$

$$(EG - 2F^2)E_y + 2EFF_y]\frac{\partial\theta}{\partial y} + H[-FE_{yy} - \frac{EF}{G}G_{yy} + 2EF_{xy} + GE_{xy} -$$

$$\frac{1}{G}(EG - 2F^2)G_{xy} - 2FF_{xy}]$$

$$+\frac{1}{H}\left[\begin{array}{l} GFE_y^2 + \frac{EF}{G^2}(2EG - F^2)G_y^2 + 4EFF_y^2 + \frac{F}{G}(EG+F^2)\times \\ E_yG_y - (EG + 3F^2)E_yF_y - \frac{E}{G}(3EG + F^2)G_yF_y - G^2E_xE_y - \\ F^2E_xG_y + 2FGE_xF_y + \frac{1}{G^2}(2H^4 - E^2G^2)G_xG_y - F^2E_yG_x + \\ 2EFG_xF_y + 2EFG_yF_x + 2GFE_yF_x - 2(EG + F^2)F_xF_y \end{array}\right]$$

$$= 0. \tag{12}$$

6. The Condition (12) When the Parametric Curves Are Orthogonal.—The parametric curves are orthogonal if and only if $F = 0$. In that event the condition (12) becomes

$$2EG\left(G\frac{\partial^2\theta}{\partial x^2} + E\frac{\partial^2\theta}{\partial y^2}\right) + G(EG_x - GE_x)\frac{\partial\theta}{\partial x} + E(GE_y - EG_y)\frac{\partial\theta}{\partial y} +$$

$$(EG)^{1/2}(GE_{xy} - EG_{xy}) + (EG)^{-1/2}(E^2G_xG_y - G^2E_xE_y) = 0, \quad (13)$$

where $\theta = \text{arc cot } p(G/E)^{1/2}$.

This is the necessary and sufficient condition that the ∞^1 curves defined by the differential equation $dy/dx = p(x, y)$ be an isothermal family when the parametric curves on the surface Σ form an orthogonal net.

Of course, if the parametric curves form an isothermal net and if x and y are isothermal parameters, then (13) reduces to Lie's theorem stating that θ is a harmonic function.

7. The Condition (12) When the Surface Σ Is Given by the Monge Equation $z = f(x, y)$.—In this case $E = 1 + f_x^2$, $F = f_xf_y$, $G = 1 + f_y^2$. Substituting these values into (12), we obtain the expression

$$(1 + f_x^2 + f_y^2)\left[(1 + f_y^2)\frac{\partial^2\theta}{\partial x^2} - 2f_xf_y\frac{\partial^2\theta}{\partial x\partial y} + (1 + f_x^2)\frac{\partial^2\theta}{\partial y^2}\right] -$$

$$\left[(1+f_y^2)f_{xx} - 2f_xf_yf_{xy} + (1+f_x^2)f_{yy}\right]\left[f_x\frac{\partial\theta}{\partial x} + f_y\frac{\partial\theta}{\partial y}\right] + \frac{f_x(1+f_x^2+f_y^2)^{1/2}}{(1+f_y^2)}\times$$

$$[(1 + f_y^2)f_{xxy} - 2f_xf_yf_{xyy} + (1 + f_x^2)f_{yyy}]$$

$$+ (1 + f_x^2 + f_y^2)^{-1/2}\left[\begin{array}{l} -4f_xf_yf_{xy}^2 - 2\dfrac{f_xf_y(1 + f_x^2)}{(1 + f_y^2)^2}(2 + f_x^2 + 2f_y^2)\times \\ \quad f_{yy}^2 + (1 - f_x^2 + f_y^2)f_{xx}f_{xy} - 2f_xf_yf_{xx}f_{yy} + \\ \quad \dfrac{1}{(1 + f_y^2)^2}\left\{\begin{array}{l} 1+2f_y^2-f_x^4+f_y^4+9f_x^2f_y^2+3f_x^4f_y^2 \\ +\ 9f_x^2f_y^4 \end{array}\right\} \\ \qquad\qquad \times f_{xy}f_{yy} \end{array}\right]$$

$$= 0, \quad (14)$$

where $\theta = \text{arc cot } [p(1 + f_y^2) + f_xf_y]/[1 + f_x^2 + f_y^2]^{1/2}$.

The preceding equation is the necessary and sufficient condition that the ∞^1 curves defined by $dy/dx = p(x, y)$ be an isothermal family on the surface Σ which is given by the Monge equation $z = f(x, y)$.

8. The Condition That the Level Curves $z = \text{Const.}$ Form an Isothermal Family.—As an application, we find that the condition that the level curves $z = \text{const.}$ of the surface Σ defined by the Monge equation $z = f(x, y)$ be

an isothermal family is given by the equation (14) where $\theta = -$ arc tan $f_y(1 + f_x^2 + f_y^2)^{1/2}/f_x$. Upon simplifying this, we find that the required third order condition is

$$(1+f_x^2+f_y^2)(f_x^2+f_y^2)[f_y(1+f_y^2)f_{xxx} - f_x(1+3f_y^2)f_{xxy} + f_y(1+3f_x^2)f_{xyy} - f_x(1+f_x^2)f_{yyy}] - 2(1+2f_x^2+2f_y^2)[(1+f_y^2)f_{xx} - 2f_xf_yf_{xy} + (1+f_x^2)f_{yy}] \times [f_xf_yf_{xx} - (f_x^2 - f_y^2)f_{xy} - f_xf_yf_{yy}] = 0. \quad (15)$$

A first integral of this equation is

$$\frac{[(1+f_y^2)f_{xx} - 2f_xf_yf_{xy} + (1+f_x^2)f_{yy}]}{(f_x^2+f_y^2)(1+f_x^2+f_y^2)} = \phi(f). \quad (16)$$

Special classes of solutions are surfaces of revolution with axes perpendicular to the xy-plane, and cylinders with elements parallel to the xy-plane. All minimal surfaces are also solutions as may be verified by (16) since for a minimal surface the numerator of the fraction vanishes. We have proved that there are no surfaces such that the isothermal system of level curves on Σ may be represented on the xy-plane by similar ellipses or hyperbolas with the same axes, or congruent parabolas with the same axis. Hence the only quadric surfaces which belong to our class are those of revolution and the cylinders.

This may be applied to the mapping upon a plane π of the loxodromes (isogonals of the level curves) of the surface Σ. By a theorem of Kasner which states that the complete system of ∞^2 isogonal trajectories of a given family is linear (in the analytic sense) if and only if the given family is isothermal, it can be shown that the loxodromes may be represented by straight lines in the plane π for a sphere (Mercator) and a spheroid (Lambert), but not for an ellipsoid of three unequal axes. Also the loxodromes can be represented by straight lines in π for any minimal surface (in any orientation) and for any surface of revolution with axis perpendicular to π.

In conclusion, we may state that the problem of this section is equivalent to the determination of the class of surfaces Σ which can be projected orthogonally upon a plane π such that the unique Tissot net (the level curves together with their orthogonal trajectories) is isothermal. This suggests for consideration the more general problem of determining the class of surfaces Σ which can be pictured by a given non-conformal transformation T upon a given surface Σ_0 such that the unique orthogonal Tissot net determined by T on Σ or Σ_0 is isothermal. A variation of this problem is to have Σ and Σ_0 given, and then to determine all transformations T with the above property.

Other generalizations and applications of our fundamental formulas will be discussed elsewhere. We show that the only surfaces which are

intersected by every set of parallel planes in an isothermal family, besides the obvious cases of spheres and and planes, are the *minimal surfaces*.

This paper was presented before the American Mathematical Society, in April, 1944.

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NEW TYPES OF RELATIONS IN FINITE FIELD THEORY

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Communicated November 27, 1944

In two other recent papers[1] we developed methods which led to several new results in finite field theory, with particular application to ordinary congruences involving rational integers. In the present article we pursue these methods much further, obtaining various new kinds of relations.

The results just referred to gave criteria involving binomial coefficients for the number of roots of an equation in a finite field. To reduce these expressions to simpler forms in order to give more convenient criteria as to the number of roots, it seems necessary to go into considerations involving binomial coefficients which have not been heretofore studied, and so far we can only use these expressions to supply this information in comparatively few cases, some of which will be discussed elsewhere. At present we shall look at the situation from another angle. It is possible to apply the criteria to certain equations where we know in advance the number of roots or some properties of them. When this is done it turns out to be a fruitful method for finding relations of an entirely new type in number theory.

As one example of this, we obtain immediately from the statement of Theorem II of the first paper[1] and the remarks just preceding it, the result that the least residue, positive or zero, of

$$A = -c^2 \sum_{r=0}^{k} \sum_{k=1}^{m} \binom{kc}{rc} a^{rc} b^{mc-kc}, \tag{1}$$

modulo p, is $\leqq c$ for p any prime of the form $1 + cm$; m odd; a and b being any integers such that $(ab, p) = 1$. In this connection we note the remarks of H. J. S. Smith[2] in connection with a congruence due to Gauss, which states that if $p = 4n + 1 = h^2 + k^2$ is a prime then

$$h \equiv \pm \frac{(2n)!}{2(n!)^2} \pmod{p}. \tag{2}$$

Smith called this a remarkable relation since h may be determined directly by finding the least residue, in absolute value, of the right-hand member and this must be also $< \sqrt{p}$. The result concerning A in (1) is somewhat analogous.

1. We now note, as did Cipolla,[3] that we may introduce functions involving the roots of a certain congruence aside from merely the number of them.

For example, consider the expression,

$$N_r = \sum (x^r - x^r f(x)^{k(p^n-1)}), \tag{3}$$

where the summation extends over all distinct elements x, $\not\equiv 0$, of a finite field $F(p^n)$. For the values x_1 of x such that $f(x_1) = 0$, this expression reduces to x_1^r, and for an x not an x_1, it vanishes in the field.

Hence

$$N_r = \sum x_1^r$$

in $F(p^n)$ where the summation extends over all distinct roots x, of $f(x) = 0$. The relation (3) gives, if $r \not\equiv 0 \pmod{p^n - 1}$ and

$$(f(x))^{k(p^n-1)} = C_0 + C + C_2x^2 + \ldots;$$

$$N_r = C_t + C_{t+s} + C_{t+2s} \ldots, \tag{4}$$

where $t = p^n - 1 - r$; $s = p^n - 1$,

and

$$N_r + 1 = C_s + C_{2s} \ldots, \tag{5}$$

for $r \equiv 0 \pmod{s}$. For the latter case N_r equals, in the field, the number of roots of $f(x) = 0$. We shall find (4) convenient for further investigations.

From the point of view we are now employing, we shall find use also for the expression

$$\sum (x^r f(x)^d - x^r f(x)^{k(p^n-1)+d}), \tag{6}$$

where the summation extends over all distinct elements $\not\equiv 0$ of $F(p^n)$ which contains (3), and $d > 0$. This function obviously reduces to zero in the field.

We now examine the relation

$$ax^m + 1 = 0; \quad p^n - 1 = mc; \tag{7}$$

and use the form (3) which gives in this case

$$N_r = \sum(x^{rm} - x^{rm}(ax^m + 1)^{k(p^n-1)}), \tag{8}$$

where the summation extends over all distinct values ρ such that $\rho^c = 1$ if we set ρ^{-1} in lieu of x^m.

Reducing (8) we obtain for $x^m = \rho^{-1}$,

$$\sum_\rho \rho^{-r} - \sum_\rho \sum_{s=0}^{k(p^n-1)} \binom{k(p^n-1)}{s} a^s \rho^{s-r}$$

or

$$-\sum_\rho \sum_{s=1}^{k(p^n-1)} \binom{k(p^n-1)}{s} a^s \rho^{s-r}.$$

Summation as to ρ gives

$$-c \sum_t \binom{k(p^n-1)}{t} a^t, \tag{9}$$

where t ranges over the integers in the set $1, 2, \ldots k(p^n - 1)$ such that $t \equiv r \pmod{c}$. We know from (7) that if $(-a)^c = 1$ then there is just one value x^m which satisfies the equation, and if $(-a)^c \not\equiv 1$ there is none, that is, $N_r = (-1)^r a^r$ or 0 in (8), for $r > 0$. Assume also $r < c$. This gives

$$c\sum \binom{k(p^n-1)}{t} a^t = (-1)^{r+1} a^r \text{ or } 0, \tag{10}$$

according as $(-a)^c = 1$ or $(-a)^c \not\equiv 1$, and if $t = 0$, then we must replace $(-1)^{r+1} a^r$ by -1 in this relation.

Now take the expression (6), and set $ax^m + 1$ for $f(x)$ and $(-r)$ for r Expansion gives, if $p^n - 1 > d > 0$,

$$c \sum_{t_1} \binom{d}{t_1} a^{t_1} - c \sum \binom{d + (p^n-1)k}{t} a^t = 0,$$

or

$$\sum_t \binom{d + (p^n-1)k}{t} a^t = \sum_{t_1} \binom{d}{t_1} a^{t_1}, \tag{11}$$

t_1 ranging over all the values in the set $0, 1, 2, \ldots, d$, such that $t_1 \equiv r \pmod{c}$, and t ranges over all the values in the set $0, 1, 2, \ldots, k(p^n - 1)$ such

that $t \equiv r \pmod{c}$. For $n = 1, a = 1, c = p^n - 1$ and $d \not\equiv 0 \pmod{p^n - 1}$ the relation (11) gives

$$\sum_t \binom{d + (p^n - 1)k}{t} \equiv \binom{d}{t} \pmod{p},$$

if $d < p^n - 1$, which is due to the writer,[4] and for $n = 1, t \equiv 0 \pmod{(p-1)}$ to Bachmann.[5]

Now set

$$\binom{v}{w} = 0,$$

for $w > v$, then relations (10) and (11) give the

THEOREM I. *If $p^n - 1 = mc$, p prime, $k > 0$, a is any element, $\neq 0$, of a finite field $F(p^n)$ then for $0 < r < c$,*

$$\sum_{s=0}^{\infty} c \binom{k(p^n - 1)}{r + cs} a^{cs} = (-1)^{r+1} a^r \text{ or } 0,$$

according as $(-a)^c = 1$, or $(-a)^c \neq 1$, also

$$\sum_{h=1}^{\infty} c \binom{k(p^n - 1)}{ch} a^{ch} = -1 \text{ or } 0,$$

with the same conditions on a.
For $d > 0, 0 \leqq r < c$, then

$$\sum_{s=0}^{\infty} \binom{d + (p^n - 1)k}{r + cs} a^{cs} \equiv \sum_{s=0}^{\infty} \binom{d}{r + cs} a^{cs}.$$

2. To obtain certain other results we shall find it convenient here to introduce in the finite field of residue classes of a prime ideal (p) where p a primitive root of a prime l and use the algebraic field defined by a primitive lth root of unity designated by ζ. The ideal (p) is a prime ideal in said field. Set $p^{l-1} - 1 = lc$. Then if a is prime to p and also $a + 1 \not\equiv 0 \pmod{p}$, then $1 + a\zeta$ is also prime to p and

$$(1 + a\zeta)^{\sigma} \equiv \zeta^k \pmod{p}, \tag{14}$$

for some k, $0 \leqq k < l$.

Expand the left hand member and collect powers of ζ, then the result may be written

$$A_0 + A_1\zeta + \ldots + A_{l-1}\zeta^{l-1} \equiv 0 \pmod{p}. \tag{15}$$

We note that this congruence also holds if we set $\zeta^2, \zeta^3 \ldots, \zeta^{l-1}$ in lieu of ζ. Then we also note that

$$A_0 + A_1 + \ldots + A_{l-1} \equiv 0, \tag{16}$$

since c is divisible by $(p-1)$ and $a+1 \not\equiv 0 \pmod{p}$. The relations (15) and (16) after making the substitutions of the various powers of ζ already indicated give l congruences from which we may eliminate the A's since the determinant formed by the ζ's is an alternant which is prime to $p \neq l$. Hence

$$A_i \equiv 0; \qquad i = 0, 1, \ldots, l-1.$$

Using the actual values of the A's we have

$$\binom{cr}{k} a^k + \binom{cr}{k+l} a^{k+l} + \ldots \equiv 1 \pmod{p},$$

and since $a \not\equiv 0 \pmod{p}$,

$$\binom{cr}{m} + \binom{cr}{km+l} a^l + \ldots \equiv 0 \pmod{p},$$

for $m \neq k$.

These give the

THEOREM II. *If p is a prime, and also a primitive root of a prime l, with $p^{l-1} - 1 = lc$, and r is any integer > 0, a is an integer with $(a(a+1), p) = 1$, then for some k in the set $0, 1, \ldots, l-1$, we have*

$$\sum_{s=0}^{\infty} \binom{cr}{k+sl} a^{k+sl} \equiv 1 \pmod{p},$$

and for any m in the set $0, 1, \ldots l-1$ with $m \neq k$, we have

$$\sum_{s=0}^{\infty} \binom{cr}{m+sl} a^{sl} \equiv 0 \pmod{p}.$$

[1] These PROCEEDINGS, **30**, 362–367, 368–370 (1944).
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FERMAT'S QUOTIENT AND RELATED ARITHMETIC FUNCTIONS

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Communicated November 27, 1944

If $(a, p) = 1$, p prime, the integer

$$q(a, p) = \frac{a^{p-1} - 1}{p} \tag{1}$$

has entered into many investigations in number theory. A number of questions concerning it are unsolved. For example, if a is fixed, is there a finite or infinite number of p's such that $q(a, p) = 0 \pmod{p}$, and if infinite is there a finite number of p's such that $a^{p-1} \equiv 1 \pmod{p^i}$ for i some given integer > 2?

Another somewhat related function is

$$W(p) = \frac{(p-1)! + 1}{p}, \tag{2}$$

known as Wilson's quotient. It differs from (1) in that it depends on p only. This function has a property[1] which connects it with the Bernoulli numbers, that is,

$$\frac{(p-1)! + 1}{p} \equiv b_{p-1} + \frac{p-1}{p} \pmod{p}, \tag{3}$$

where $(b + 1)^n = b_n$; $n > 1$, and after expansion we set $b^k = b_k$. Our object here is to introduce two other arithmetic functions (designated by G and M/p below) expressed as quotients, each of which is related to Fermat's quotient or the Bernoulli numbers.

1. We know that $b_k = 0$ for $k > 1$ and odd. Set $b_{2n} = (-1)^{n-1} B_n$ and further, write

$$B'_n = \frac{B_n}{n},$$

then consider the known relations

$$B'_{n+\mu} - B'_n \equiv 0 \pmod{p}; \quad \mu = (p-1)/2. \tag{4}$$

We then define the function

$$G(n, p) = \frac{B'_{n+\mu} - B'_n}{p}. \tag{5}$$

We have

$$G(n, p) \equiv G(n + \mu, p) \pmod{p},$$

so that there are only $p - 1$ incongruent G's modulo p. This follows from the relation[2]

$$B'_{n+y\mu} = yB'_{n+\mu} - (y - 1)B'_n.$$

We also infer from this that we get no essentially new functions by using $(B'_{n+k\mu} - B'_{n+s\mu})$ instead of $B'_{n+\mu} - B'_n$. We shall be interested here in the problem of transforming $G(n, p)$, modulo p, with $B_n \equiv 0 \pmod{p}$. There is no known case[2] where $G(n, p) \equiv 0 \pmod{p}$ under this restriction.

We have[3]

$$\frac{n^{2i} - 1}{2i} b_{2i} \equiv \sum_{a=1}^{p^2-1} y_a a^{2i-1} \pmod{p^2},$$

and

$$\frac{n^{2i+p-1} - 1}{2i + p - 1} b_{2i+p-1} \equiv \sum_{a=1}^{p^2-1} y_a a^{2i+p-2} \pmod{p^2},$$

and for $b_{2i} \equiv b_{2i+p-1} \equiv \pmod{p}$; $\quad i > 1$

$$\frac{n^{2i} - 1}{2i(2i - 1)} G(2i, p) \equiv \sum y_d d^{2i-1} q(d) \pmod{p}, \qquad (6)$$

the summation extending over all values d in the set $1, 2, \ldots, p^2 - 1$ which are prime to p.

2. In another paper[4] the writer founded an arithmetical theory of the Bernoulli numbers on the congruence

$$S_n = 1^n + 2^n + \ldots + (p - 1)^n \equiv pb_n \pmod{p^2}$$

$n < p - 1$. Now S_n has the property that it is divisible by p for $n < p-1$. So also has the function

$$A = \sum_a a^n$$

where a ranges over all the positive integers, $a < (p - 1)$ such that $a^c \equiv 1 \pmod{p}$; also, $n \not\equiv 0 \pmod{c}$. However, all proofs of the above property of S_n seem to depend on additive properties of this function, whereas $\sum_a a$ is defined by means of multiplicative properties of certain integers and many of the additive properties of S_n and all apparently fail to carry over to A. Nevertheless, there is a relation between A/p and the Bernoulli numbers, and it is given in Theorem I below. This was a quite unexpected result. Consider the expression

$$\sum_\rho (1 - (a\rho + 1)^{p-1})^2 \equiv N \pmod{p^2}, \qquad (7)$$

where ρ ranges over all the values such that $\rho^c \equiv 1 \pmod{p^2}$, where $p - 1 = mc$. Any term in this expression reduces to unity, modulo p^2, if ρ is such that

$$a\rho + 1 \equiv 0 \pmod{p}, \tag{8}$$

for $p > 2$. If this is not the case, however, then for such a value of the corresponding term (7) is divisible by p^2 Hence, N in (7) is the number of incongruent solutions ρ, modulo p, in (8). The number of such incongruent solutions is 1 or 0, according as $(-a)^c \equiv 1 \pmod{p}$ or $(-a)^c \not\equiv 1 \pmod{p}$. Expansion of (7) gives

$$N \equiv c - 2c \sum_{k=0}^{m} a^{kc} \binom{p-1}{kc} + c \sum_{k=0}^{2m} a^{kc} \binom{2(p-1)}{kc},$$

modulo p^2, and this may be written

$$N \equiv -2c \sum_{k=1}^{m} a^{kc} \binom{p-1}{kc} + c \sum_{k=1}^{m} a^{kc} \binom{2(p-1)}{kc} + c \sum_{t=1}^{m} a^{c(m+t)} \binom{2(p-1)}{(m+t)c}.$$

Multiply through by a^n, $n \not\equiv 0 \pmod{c}$, and using the value of N noted above, we have, after letting a range over the integers $1, 2, \ldots, p-1$ and adding, we obtain

$$\sum r^n \equiv -2c \sum_{k=1}^{m} S_{kc+n} \binom{p-1}{kc} + c \sum_{k=1}^{m} S_{kc+n} \binom{2(p-1)}{kc} + c \sum_{k=1}^{m} S_{cm+ck+n} \binom{2(p-1)}{cm+ck},$$

modulo p^2, where

$$S_i = 1^i + 2^i + \ldots + (p-1)^i,$$

and r ranges over the integers in the set $1, 2, \ldots, p-1$, such that $(-r)$ satisfies $x^c \equiv 1 \pmod{p}$. Using the formula, for i even,

$$S_i \equiv pb_i \pmod{p^2}, \tag{10}$$

and dividing (9) through by p, we obtain, modulo p, with c and n even,

$$\frac{\sum r^n}{p} \equiv -2c \sum_{k=1}^{m} \binom{p-1}{kc} b_{kc+n} + c \sum_{k=1}^{m} \binom{2(p-1)}{kc} b_{kc+n} + c \sum_{k=1}^{m} b_{ck+cm+n} \binom{2(p-1)}{cm+ck}. \tag{11}$$

It is known[5] that, modulo p,

$$\binom{\alpha p + \beta}{\alpha_1 p + \beta_1} \equiv \binom{\alpha}{\alpha_1}\binom{\beta}{\beta_1}$$

each of the integers α, β, α_1, β_1, being $\geqq 0$ and $< p$.
Employing this, we note that, modulo p,

$$\binom{2(p-1)}{kc} \equiv \binom{p+p-2}{kc} \equiv \binom{p-2}{kc},$$

and

$$\binom{2(p-1)}{p-1+tc} \equiv \binom{p+p-2}{p+tc-1} \equiv \binom{p-2}{tc-1}.$$

Hence (11) reduces, modulo p, to

$$-2c\sum_{k=1}^{m} b_{kc+n}(-1)^{kc} + c\sum_{k=1}^{m} b_{kc+n}\binom{p-2}{kc} + c\sum_{t=1}^{m}\binom{p-2}{tc-1}b_{cm+ct+n}. \quad (12)$$

It is also known that

$$\binom{p-1}{r} \equiv (-1)^r \pmod{p}.$$

We also have

$$\binom{p-2}{kc} = \frac{(p-2)(p-3)\ldots(p-1-kc)}{2.3.\ldots kc}.$$

Now

$$(p-2)(p-3)\ldots(p-(1+kc)) \equiv (-1)^{kc}\,2, 3, \ldots (kc+1).$$

so that

$$\binom{p-2}{kc} \equiv (-1)^{kc}(kc+1) \pmod{p}.$$

Also

$$\binom{p-2}{tc-1} = \frac{(p-2)(p-3)\ldots(p-tc)}{2.3\ldots(tc-1)}$$

$$\equiv (-1)^{tc-1}\,tc \pmod{p},$$

hence (12) becomes

$$-2c\sum_{k=1}^{m} b_{kc+n}(-1)^{kc} + c\sum_{k=1}^{m} b_{kc+n}(-1)^{kc}(kc+1) + c\sum_{t=1}^{m}\binom{p-2}{tc-1} b_{p-1+ct+n}. \quad (13)$$

Now use the relation

$$\frac{b_{s+p-1}}{s+p-1} \equiv \frac{b_s}{s} \pmod{p}$$

for $s \not\equiv 0 \pmod{p-1}$, we may then write, modulo p, if $M'(c, p, n)$ is $\sum_r r$ where r ranges over the integers h such that $(-h)^c \equiv 1 \pmod{p}$.

$$\frac{M'(c,p,n)}{p} \equiv c\sum_{k=1}^{m}(-1)^{kc}(kc+1)\,b_{kc+n} + c(-1)^{kc-1}\sum_{k=1}^{m} kc\frac{kc+n-1}{kc+n} b_{kc+n} - 2c\sum_{k=1}^{m} b_{kc+n}(-1)^{kc}.$$

and collecting the terms on the right we find

$$\frac{M'(c,p,n)}{p} \equiv cn\sum_{k=1}^{m}(-1)^{kc-1}\frac{b_{kc+n}}{kc+n}, \quad (14)$$

modulo p.

Now, since c and n are even,

$$\frac{M'(c,p,n)}{p} \equiv \frac{M(c,p,n)}{p} \pmod{p},$$

where $M = \sum_a r^n$ where r ranges over the incongruent integers $< p$ which satisfy $x^c = 1$, hence we may simplify (14) and obtain

THEOREM I. *If p is an odd prime with $p - 1 = mc$, and $M(c, n, p)$ is defined by*

$$\frac{\sum r^n}{p},$$

where the summation extends over all integers r in the set $1, 2, \ldots, p-1$ such that $r^c \equiv 1 \pmod{p}$, with $n \not\equiv 0 \pmod{c}$, then if c and n are even,

$$M(c,n,p) \equiv -cn\sum_{k=0}^{m-1}\frac{b_{kc+n}}{kc+n} \pmod{p}. \quad (15)$$

For $c = p - 1$, then $m = 1$ and the result becomes the well-known relation (10).

It is possible to extend the Theorem I to the sum

$$\sum \frac{(ma + k)^n}{p},$$

where a ranges over the integers in the set $1, 2, \ldots, p - 1$ such that $a^c \equiv 1 \pmod{p}$, and so that the b's are replaced by numbers of the form $(mb + h)^{kc+n}$, but the proof will not be given here. Also it follows from Theorem I that

$$M(c, n, p)/n \equiv M(c, n + c, p)/(n + c) \pmod{p}.$$

[1] A proof is given by the writer, as a special case of an explicit expression for any Bernoulli number, in *Duke Math. Jour.*, **8**, 578 (1941), relation (26).

[2] Pollaczek, *Math. Zeit.*, **21**, 36 (1924).

[3] Vandiver, *Duke Math. Jour.*, **5**, 549 (1939).

[4] *Trans. Amer. Math. Soc.*, **51**, 510 (1942).

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NON-LINEAR INTEGRAL EQUATIONS OF THE HAMMERSTEIN TYPE

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Communicated November 14, 1944

Sufficient conditions for the existence and uniqueness of a solution to the integral equation:

$$\psi(x) = \int_a^b K(x, y) f\{y, \psi(y)\} dy \qquad (1)$$

were investigated by Hammerstein[1] and his pupils. If $K(x, y)$ is a sym metric, positive-definite continuous kernel* in L^2 and $f(x, y)$ is continuous in x and y, their results may be classified into three closely related categories, according to the degree of non-linearity permitted $f(x, y)$

Hammerstein obtained the most general theorem by requiring that (x, y) satisfy the inequality:

$$\int^v f(x, y) dy \leqslant \mu_1 \frac{v^2}{2} + C_1; \quad C_1 > 0; \quad 0 < \mu_1 < \lambda_1 \qquad (A)$$

for all v. Here λ_1 is the smallest characteristic value of $K(x, y)$ in the sense of linear integral equations. Under this condition Hammerstein established the existence of at least one function $\psi(x)$ satisfying (1). His method was essentially that of the Rayleigh-Ritz process, (A) being sufficient to guarantee the existence of a lower bound to the functional possessing (1) as its Euler-Lagrange equation.

Iglisch[2] imposed a stronger condition on $f(x, y)$; namely,

$$0 < \lim_{|y| \to \infty} \operatorname{Sup} \frac{f(x, y)}{y} < \mu_1 < \lambda_1. \qquad \text{(B)}$$

Utilizing reasoning similar to that of the fixed-point method of Schauder-Leray, Iglisch proved the existence of a solution under (B).

Both Hammerstein and Iglisch also treated (1) under a still stronger condition on $f(x, y)$. In the event that $f(x, y)$ satisfied the inequality:

$$\left| \frac{f(x, y_2) - f(x, y_1)}{y_2 - y_1} \right| \leqslant \mu_1 < \lambda_1 \text{ for any } y_1, y_2, \qquad \text{(C)}$$

both men were able to establish the existence and uniqueness of a solution to (1). Golomb[3] also applied the Picard Approximation Process to construct a solution under (C) even when $K(x, y)$ was unsymmetric and admitted an absolutely and uniformly convergent bilinear expansion in its characteristic functions.

In the event that $f(x, \psi)$ is a linear function in ψ, the conditions (A), (B) and (C) are all equivalent and the above results are classical. As is well known, in the linear theory it is not essential to limit the above inequalities to the smallest characteristic value. In fact all that is required is that the linear function f be bounded from above and below by consecutive characteristic values λ_n, λ_{n+1}. It is therefore natural to consider the non-linear integral equation (1) under the corresponding more general conditions: for all v

$$\mu_n \frac{v^2}{2} + Cn \leqslant \int_0^v f(x, y)\, dy \leqslant \mu_{n+1} \frac{v^2}{2} + C_{n+1} \qquad \text{(A')}$$

$$0 < \lambda_n < \mu_n < \mu_{n+1} < \lambda_{n+1}$$

$$\mu_n \leqslant \liminf_{|y| \to \infty} \frac{f(x, y)}{y} \leqslant \limsup_{|y| \to \infty} \frac{f(x, y)}{y} \leqslant \mu_{n+1} \qquad \text{(B')}$$

$$\mu_n \leqslant \left| \frac{f(x, y_2) - f(x, y_1)}{y_2 - y_1} \right| \leqslant \mu_{n+1} \qquad \text{(C')}$$

for any y_2, y_1.

In the cases (B') and (C') the theorem corresponding to (B) and (C) and their methods of proof can be readily generalized. These proofs depend upon the existence of an *a priori* estimate of the norm of all possible solutions to (1) under the above conditions of $f(x, y)$. In order to establish an estimate, use was made of the fact that (B'), and hence *a fortiori* (C'), imply that $f(x, y)$ can be written as:

$$f(x, y) = \left(\frac{\mu_{n+1} + \mu_n}{2}\right)y + g(x, y) \tag{2}$$

where for sufficiently large N, $|y| > N$

$$|g(x, y)| < {}^1/_2\{\mu_{n+1} - \mu_n\}|y|. \tag{3}$$

This enables (1) to be written as one of the equations of the family defined by:

$$\psi_\tau(x) = \frac{(\mu_{n+1} + \mu_n)}{2} \int_a^b K(x, y)\psi_\tau(y)dy + \tau \int_a^b K(x, y)g[y, \psi_\tau(y)]dy. \tag{4}$$

That is, for $\tau = 1$, equation (4) reduces to (1); and for $\tau = 0$, equation (4) is an ordinary Fredholm equation of the first kind, which, since $\frac{\mu_{n+1} + \mu_n}{2}$ is distinct from all the characteristic values of $K(x, y)$, possesses only the solution $\psi_0(x) = 0$. Moreover, for any value of τ,

$$||\psi_\tau||^2 \leqslant \frac{A^2}{\delta^2 - \frac{\tau}{2}[\mu_{n+1} - \mu_n]^2} \leqslant \frac{A^2}{\delta^2 - \frac{[\mu_{n+1} - \mu_n]^2}{2}} \tag{5}$$

where

$$||g(\psi_\tau)|| \leqslant A \text{ when } ||\psi_\tau|| < N \tag{6}$$

and

$$\delta^2 = \text{Min}\left\{\left[\lambda_{n+1} - \frac{(\mu_{n+1} + \mu_n)}{2}\right]^2, \left[\lambda_n - \frac{(\mu_{n+1} + \mu_n)}{2}\right]^2\right\}. \tag{7}$$

The fixed-point method of Schauder-Leray[4] can now be applied and the existence of a solution concluded under (B′) almost immediately. The estimate given by (5) is also entirely sufficient to prove the uniqueness of the solution under (C′).

In distinct contrast to this, no scheme yielded an *a priori* estimate under (A′). In fact, it is an easy matter to construct an example demonstrating that there is no topological reason for such an estimate to exist under these conditions. Thus the local methods of the calculus of variations have to be replaced by those in the large before further progress is possible. The generalization of (A) which was finally achieved is very incomplete, even though it does employ the methods in the large in a somewhat different way than they had hitherto been used. The method of proof does not in any way depend upon the existence of an *a priori* estimate, although, remarkably enough, once the existence of a solution has been established, it does permit an estimate of the norm of the solution to be made (*a posteriori*

estimation). Before stating these results and sketching the method of proof, it is convenient to state that equation (1) is equivalent to

$$\phi(x) = \int_a^b H(x, y) f \{y \int_a^b H(y, s) \psi(s) ds\} dy \tag{8}$$

where

$$K(x, y) = \int_a^b H(x, s) H(s, y) ds \tag{9}$$

in that if $\psi(x)$ is a solution of (1), then

$$\phi(x) = \int_a^b H(x, y) f [y, \psi(y)] dy \tag{10}$$

is a solution of (B), and conversely, if $\phi(x)$ is a solution of (8), then

$$\psi(x) = \int_a^b H(x, y) \phi(y) dy \tag{11}$$

is a solution of (1). Furthermore, equation (8) is the Euler-Lagrange equation of the functional

$$J(\phi) = (\phi, \phi) - 2G(H\phi) \tag{12}$$

where

$$G(H\phi) = \int_a^b x \int_0^{\int_a^b H(x, y)\phi(y)dy} f(x, v) dv \tag{13}$$

THEOREM: If $f(x, y)$ satisfies

$$\mu_n(\phi, \phi) - C_n \leqslant G(\phi) \leqslant \mu_{n+1}(\phi, \phi) + C_{n+1} \qquad (A'')\dagger$$

where μ_n, μ_{n+1} are less the nth and $n+1$st characteristic values of $K(x, y)$ respectively; if

$$|f(x, y)| < A |y| + B \tag{14}$$

for some constants $A > 0$, $B > 0$; and if the functional $J(\phi)$ possesses only one maximum on any n-dimensional linear manifold in Hilbert space parallel to the one containing the origin and the first n characteristic functions of $K(x, y)$; then equation (1) has at least one solution.

Sketch of the Proof: Let Γ denote the class $\{M_n\}$ of n-dimensional manifold described above, M_n^0 the particular one containing the origin. Define

$$d(M_n) = \operatorname{Sup}_{\phi \in M_n} J(\phi) \tag{15}$$

and

$$d = \inf_{M_n \in \Gamma} d(M_n). \tag{16}$$

Condition (A'') implies that the number d defined by (16) is finite. Each M_n^a is uniquely defined by its intersection with the manifold orthogonal

to M_n^0. This intersection will be denoted by ψ^α. For all manifolds such that

$$d(M_n^\alpha) < d(M_n^0) \tag{17}$$

the norm of the function ψ^α is governed by the inequality

$$||\psi^\alpha|| < G(M_n^0). \tag{18}$$

Thus, if a sequence of manifolds $\{M_n^\beta\}$ is chosen so that

$$d(M_n^\beta) \to d \tag{19}$$

the corresponding sequence $\{\psi^\beta\}$ will converge to a value, ψ^* since any bounded set of Hilbert space is sequentially compact in the usual weak topology [in which neighborhoods are defined in terms of an ϵ and any finite set of linear functions**].

The linear manifold M_n^* containing ψ^* and parallel to M_n^0 is a limit manifold for which

$$d(M_n^*) = d \tag{20}$$

and on which, by hypothesis, there exists a point ϕ_0 such that

$$J(\phi_0) = d. \tag{21}$$

Furthermore, unless

$$\text{grad } J(\phi_0) = 0 \tag{22}$$

it would be possible to displace M_n^* into a new manifold M_n^{**} on which

$$d(M_n^{**}) < d \tag{23}$$

contrary to the definition of d. One merely has to consider the displacement defined by

$$\phi_\tau = \phi_0 + \tau \text{ grad } J(\phi_0) \tag{24}$$

and to divide M_n^* into three concentric regions located around ϕ_0 as center. The first of these is defined as the region of ϕ_0 where grad $J(\phi)$ is uniformly bounded away from zero, the third as the region at infinity where

$$J(\phi) < \frac{d}{2} \tag{25}$$

and the second as the difference between the first and the third. The existence of the third region follows directly from the original hypotheses. The problem then reduces to a finite dimensional problem in n-variables and it follows by an examination of all possible cases that (24) would permit M_n^* to be displaced into M_n^{**}, satisfying (23). From this contradiction and (8) and (11), it follows that equation (1) has at least one solution. Moreover, since

$$||\psi^*|| < G(M_n^0) \tag{26}$$

and

$$J(\phi_0) = d, \phi_0 = \theta + \psi^* \tag{27}$$

an estimate of θ, and hence of ϕ_0, can be made with the aid of (A″).

In the event that $f(x, y)$ is linear and of the form

$$f(\phi) = \lambda\phi(x) + h(x) \tag{28}$$

this method of proof also yields an interesting geometrical interpretation of the relationship between the homogeneous and nonhomogeneous linear Fredholm problems which agrees with the one first given by Lusternik and Schnirelman[5] for the finite-dimensional analogue of the homogeneous case. It can be shown that the usual characteristic functions and values are given by considering linear cycles in the projective Hilbert space, which may be thought of as lying on a sphere at infinity with its diametrical opposite points identified and that the solutions to the non-homogeneous problem for values of λ between successive characteristic values may be obtained as above by considering n-dimensional linear manifolds passing through Hilbert space and parallel to M_n^0.

The complete and detailed proofs of the above results will be published in a forthcoming paper.

* The theorems are still true if the kernels are only "brauchbar unstetig."

† (A″) on equation (8) is easily seen to be the same as (A′) for equation (1).

** See, e.g., F. H. Murray, *Linear Transformations in Hilbert Space*, Princeton University Series.

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